

Surveillance

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INSIDE:

Surveillance and intelligence – working together
Quarterly report of investigations of suspected exotic diseases
Quarterly report of investigations of suspected exotic marine
and freshwater pests and diseases

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Editorial

Surveillance and intelligence – working together

Surveillance is an essential component of our biosecurity system. By monitoring risk offshore, at border crossing points and in the domestic environment, we gain the earliest possible indication that a risk has changed or arrived in New Zealand.

The Ministry for Primary Industries (MPI) is always trying to find ways to better integrate the components of the biosecurity and surveillance systems because coherent, targeted surveillance gives us options. When we understand risk, or have systems in place to alert us, we can choose where to allocate different intervention tools, including increased levels of vigilance, border actions or investigations.

One way of progressing this change is how we use the intelligence and targeting system. Intelligence is part of the Intelligence Planning and Coordination Services Directorate, which supports biosecurity by providing actionable insights, planning and development support, operational co-ordination and operational support services. We are in the process of evolving our intelligence processes, so that we integrate better with other components of the surveillance system, provide more usable outputs, and support a more responsive biosecurity system.

The first part of this journey has been to make better use of the biosecurity data already available to us. To help all border agencies generate more value from their border data, MPI, Customs and the Ministry of Business, Innovation and Employment recently established a Joint Border Analytics Centre in Auckland. This team uses predictive modelling to understand border risk, and is driving a lot of conversations about how we can integrate different data sets to understand our system better, and learn from each other in the field of data analytics.

The first project undertaken by MPI staff in this new capability was an analysis of interceptions on fresh produce and cut flowers imports. There is a lot of focus on these pathways because of the risk that infestation would pose to our domestic and export industries. We need to know when there are high-risk interceptions, but also how well the pathway is performing overall and the key risk drivers that we could address with various interventions. The outputs from this team and the Intelligence and Targeting Team will in the future help prioritise offshore pathway assurance activities, target border inspections and help us understand which commodities and pathways are performing well.

One of the main challenges when undertaking this analysis is the data. Interception data is stored in different databases, with many different fields and characteristics. Using data analytics gives us the ability to bring together the outputs of these data sets without requiring significant changes in how we collect or structure data. It also helps us prioritise where small

enhancements in data collection practice would increase our accuracy and effectiveness, thereby helping us with continuous improvement. We are lucky to be working with a wide analytics community, including the Centre of Excellence for Biosecurity Risk Analysis (CEBRA) and Department of Agriculture and Water Resources (Australia) so that we can learn from previous experience.

We are also trying to compare risk in different pathways so we can move resources around the country. If you'll excuse the pun, it's a bit like comparing apples and oranges! But it's work that needs doing if we are to ensure that we allocate resource to areas of higher risk. For example, last summer we used intelligence assessments to help us reduce interventions in the cruise pathway by 27 percent, and we instead focused on checking that appropriate procurement processes, surveillance and pest management systems were in place on cruise vessels. This meant that we could release quarantine officers, dogs and X-ray equipment back into high-risk areas like inspections for brown marmorated stink bug (BMSB).

Throughout the summer we also monitored BMSB border interceptions, offshore distribution and actions taken by shipping companies and our equivalent agencies overseas. This included exchanging interception data with Australia to see if the patterns they were seeing were similar (they were!). This enabled us to adjust our alerts and inspections regime where necessary, so that we could monitor high-risk commodities and companies. We also provided monthly reports on BMSB interceptions and offshore activities to MPI teams, including post-border surveillance, so that they could prioritise their monitoring and responses.

Another important step has been actively increasing the amount of information that we share with industry partners and stakeholders. Under its Biosecurity 2025 strategy, MPI shares management of the biosecurity system with stakeholders across the country. This is something we need to keep working at, in order to ensure that we share the maximum possible while also maintaining our role as regulators. We know that it is important to put information in the hands of all those who contribute to the biosecurity system so that we can all choose actions that better protect our environment and industries.

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ANIMALS

Quarterly report of diagnostic cases: January to March 2017

SVS Laboratories

BOVINE

A suspected drench-gun injury in an R1 steer in the Waikato district caused a chronic inspissated abscess in the nasopharynx, leading to dyspnoea and dysphagia. Smears of the tissue submitted were negative for acid-fast bacteria but revealed Gram-positive organisms. Cultures isolated *Trueperella pyogenes*.

Two 3-year-old Jersey cows from a 400-cow dairy herd in the Tasman district presented with diarrhoea and weight loss. Serology tests for *Mycobacterium avium* ssp. *paratuberculosis* were positive, consistent with a diagnosis of **Johne's disease**.

Four 5-month-old dairy calves in a group of 30 in the Matamata district had diarrhoea and weight loss. Culture of faecal samples yielded a moderate growth of *Yersinia pseudotuberculosis* and a lighter growth of *Campylobacter jejuni*. A diagnosis of **yersiniosis** was made. This is not commonly seen during the summer months, but was a likely consequence of the wetter conditions this season.

Three cows aborted at about 8 months' gestation on a South Waikato dairy farm, prompting submission of one aborted fetus. Histopathology of multiple fetal tissues revealed lesions typical of *Neospora caninum*. In addition, PCR tests on fetal stomach contents were positive for both *N. caninum* and *Aspergillus fumigatus*, both considered to be significant (although the placenta was not available to confirm a mycotic placentitis) and giving multiple aetiologies of **neosporosis** and **mycotic abortion**. An immunologic stressor such as this mycotic infection may have caused re-activation of a latent neospora infection in the dam.

Mycotic abortion was diagnosed after fetal histopathology in another case of bovine abortion. Histopathology of the liver and adrenal gland revealed inflammatory aggregates in both, and branching fungal hyphae were seen in PAS-stained sections.

Two South Waikato dairy heifers presented with similar peri-anal protruding masses. Resected tissue from one was submitted for histopathology and revealed a low grade **soft-tissue sarcoma**. The involvement of bovine papillomavirus (BPV) was suspected. The farmer reported that shrinkage of the masses occurred in both heifers, which is the usual outcome of BPV infection in normal, healthy animals. PCR and DNA sequencing carried out at Massey University confirmed the underlying aetiology as **bovine papillomavirus type 2**.

The sudden death of two 6-month-old calves in the Rotorua district, the only calves that had been grazing in a paddock topdressed with a nitrogen fertiliser, was investigated. The calves were in good body condition and had been vaccinated with clostridial and leptospiral vaccines. Fixed necropsy tissues from one calf were submitted. The most significant findings were seen in the lung, where there were marked pulmonary interstitial and alveolar oedema and congestion, alveolar hyaline membranes and alveolar type 2 hyperplasia. These findings were consistent with damage caused by a pneumotoxic agent, and a presumptive diagnosis of **3-methylindole (3MI) toxicity (fog fever)** was made, consistent with the grazing history. This respiratory condition is most often seen in late summer and early autumn when cattle are moved onto lush pasture (particularly paddocks that have recently received an application of nitrogenous fertiliser), causing a sudden change in dietary pattern that disrupts the normal equilibrium of the ruminal microflora. The increased dietary protein (specifically the amino acid L-tryptophan) provides a substrate for a two-step conversion to 3MI by rumen microbes. On reaching the pulmonary tissues via the bloodstream, 3MI is metabolically activated by mixed-function oxidases in the bronchiolar club cells and alveolar type 1 pneumocytes releasing pneumotoxic metabolites, notably free radicals (Bray *et al.*, 1993). The young age of the calves affected in this case was

noteworthy, because the condition is more often seen in adult cattle.

Several cases of **zinc toxicity** were seen in dairy cattle, caused by overdosing of zinc administered to prevent facial eczema (FE). In one case, a South Waikato dairy cow presented with red urine, pale mucous membranes and tachycardia. Serum zinc was markedly increased, at 210 $\mu\text{mol/L}$ (reference range 11–20; prophylactic range for FE prevention 20–35), and this was consistent with **zinc toxicity**. Haematology revealed that the RBCs were at the low end of normal ($5.7 \times 10^{12}/\text{L}$; reference range $5.0\text{--}7.7 \times 10^{12}$) and Heinz bodies were elevated, at 62 percent (reference range < 1). This indicated oxidative damage, the initiating cause of the ensuing intravascular haemolysis; hence the red urine was due to haemoglobinuria, which in itself causes further renal pathology.

Another Waikato dairy cow presented clinically with pale mucous membranes, lethargy and profuse, foul-smelling diarrhoea. Haematology confirmed severe anaemia (HCT 0.09; reference range 0.24–0.40), which was markedly regenerative (reticulocytes $165.6 \times 10^9/\text{L}$; reference range $< 1 \times 10^9$). These results suggested haemolysis, a diagnosis that was backed up by high serum bilirubin (56.1 $\mu\text{mol/L}$; reference range 0.0–13.0). Serum zinc was elevated, at 48 $\mu\text{mol/L}$ (reference range 11–20 and 20–35 for FE prevention), and a presumptive diagnosis of **zinc toxicity** was made. Acute ingestion of highly concentrated zinc solutions causes severe gastroenteritis, which would account for the severe diarrhoea.

A 6-month-old dairy/beef bull calf presented initially with pale mucous membranes and deteriorated rapidly. Bloods submitted revealed a moderately regenerative, marked anaemia (HCT 0.08; reference range 0.23–0.42), together with increased bilirubin (39.7 $\mu\text{mol/L}$; reference range 0–13), consistent with haemolysis. No *Theileria* organisms were seen on the smear (although PCR was not done to confirm their absence) but serum zinc was found to be excessive,

at 47 $\mu\text{mol/L}$ (reference range 11–20 and 20–35 for FE prevention), and a presumptive diagnosis of **zinc toxicity** was made. Again, the serum level was lower than sometimes associated with haemolysis induced by excess zinc, but susceptibility is greater in younger animals like this. The calf was euthanased *in extremis* and tissues submitted. Histologic changes in the kidney were consistent with haemoglobinuric nephrosis. Other causes of haemolytic anaemia were ruled out by the clinical history.

Theileria sp. organisms were seen in a blood smear from an anaemic Waikato dairy cow (HCT 0.08; reference range 0.24–0.40 and haemoglobin 21 g/L; reference range 85–130), giving a presumptive diagnosis of **theileriosis**.

Salmonella Typhimurium phage type 108/170 was isolated from a faecal sample from a 4-day-old Wagyu calf with diarrhoea. The calf also had a moderate infection of **cryptosporidia**. The farm was reported to have had a high incidence of calf diarrhoea in the previous calving season.

Two 6-month-old hand-reared lambs in the Bay of Plenty died after a short illness depicted by abdominal pain. Necropsy of one lamb revealed icteric carcass fat and liver, serosanguinous fluid in body cavities, and black kidneys. Histopathology revealed hepatic cholestasis and periportal bridging fibrosis, consistent with **chronic copper toxicity**. The mineralised supplementary pellets that the lambs had received were the most likely copper source.

EQUINE

Cultures of a swab from a submandibular abscess in a two-year old Waikato Thoroughbred filly grew **Streptococcus equi ssp. zooepidemicus**. Although this is often considered an opportunist pathogen, recent work (Patty *et al.*, 2016) reveals that genetic variation in virulence factors determines the organism's ability to attach to equine tissues, and therefore its pathogenicity. As a result, some strains appear to be the primary pathogen in equine respiratory and uterine infections.

In another case of submandibular lymph-node swelling, a swab from the lymph node of a 5-month-old Thoroughbred filly in the Waikato was positive on PCR for **Streptococcus equi ssp. equi**, confirming a diagnosis of **strangles**.

AVIAN

An eight-month-old free-range California quail cock, part of a collection of game birds on the property, had a short history of lethargy and crop distension. It later died and the carcass was submitted for examination. Histological evaluation of necropsy tissues revealed a heavy infestation of **Capillaria sp.** within the crop. There was chronic mucosal inflammation in the crop, oesophagus and proventriculus, together with secondary bacterial infection and evidence of hepatic bacterial dissemination. A similar problem had arisen the previous year in some of the game birds kept in cages on the ground, but this had resolved with elevation of the cages.

CANINE

A 9-year-old female Labrador-Poodle crossbred dog in the Hamilton area presented with a history of coughing. She showed a poor response to antibiotics, then an initial good response to glucocorticoid therapy, but later relapsed. Radiographs revealed a large mass in the caudal lung lobe, which was resected and submitted. On histopathological examination a **metastatic malignant pilomatricoma** was diagnosed. This is a rare tumour in dogs and originates in dermal follicles. Further investigation revealed that a skin mass on the dog's left forepaw, excised 3 years previously, may have been the primary tumour, though histology had not been done to confirm this.

FELINE

A 12-year-old neutered female Burmese cat that initially presented with polyuria/polydipsia had an in-clinic blood glucose level of 24.8 mmol/L (reference range 3.6–6.3) and glucosuria on urine dipstick, consistent with **diabetes mellitus**. Because the cat's condition was worsening, blood samples were submitted for further chemistry. Serum fructosamine was elevated, at 615 $\mu\text{mol/L}$ (reference range 190–365), confirming persistent hyperglycaemia together with a metabolic acidosis (bicarbonate 13.6 mmol/l; reference range 17–24) and an anion gap of 42 mmol/L (reference range 18–25) as a result of **severe ketoacidosis**. This was confirmed by serum beta-OH butyrate analysis (uncommonly assessed in cats), which was markedly increased to 10.53 mmol/L (reference range < 0.11).

New Zealand Veterinary Pathology

BOVINE

A seven-month-old calf on a dairy property in the Waikato had diarrhoea. On clinical examination the animal appeared dehydrated and had a temperature of 39.5°C. The next morning it was found dead and a postmortem was performed, which revealed an oedematous large intestine. The intestinal contents were watery and blood-tinged. Histology revealed the presence of numerous intranuclear inclusion bodies in the vascular endothelium of the kidney and intestine, along with foci of necrosis within the lymphoid follicles of the Peyer's patches. This was consistent with infection by **bovine adenovirus**, which is a common infection although many cases are subclinical. Mildly infected animals may exhibit only a mild ocular and/or nasal discharge, but more severe cases have respiratory distress and/or diarrhoea with underlying immunocompromise, including bovine viral diarrhoea (BVD). Unfortunately, no further testing for BVD virus was undertaken in this case.

An eight-year-old cow in the Matamata-Piako district appeared to go blind over a period of about two weeks and also developed depression and deafness. The cow was otherwise clinically normal except for a mildly increased temperature. The cow was euthanased and the brain was submitted to the laboratory under the MPI transmissible spongiform encephalopathy (TSE) surveillance programme. Histology revealed a pituitary chromophobe neoplasm with a high mitotic index but no evidence of invasion. The lack of local invasion favours a diagnosis of **pituitary chromophobe adenoma**, a very rare neoplasm in cattle.

A mature dairy cow in the Western Bay of Plenty district died suddenly and had a bloody discharge present at the nose and anus. There was a small amount of bloody serum in the chest and abdomen. On histology, numerous septic emboli were seen distributed throughout the lungs. While no other sites of infection were identified grossly or histologically, it is likely that these septic emboli were the result of **vena caval syndrome**, in which a liver abscess erodes through the wall of a vessel draining into the vena cava, resulting in the showering of the lungs by septic emboli.

A yearling Angus bull in the Waipa district was a member of a small group of bulls (mostly from the same sire), several of which had developed severe lameness. This animal was slaughtered at home and the skinned forelimb was submitted after butchering for examination of the shoulder joint. The joint cavity appeared swollen. The glenoid cavity showed almost complete erosion of the cartilage affecting half of the cavity, and roughened subchondral bone was exposed. The articular surface of the humerus was markedly flattened, with a central region of cartilage erosion and marked wearing of the articular surface. Numerous small fragments of bony tissue (joint mice) were palpable in soft connective tissues surrounding the joint. Severe **degenerative joint disease secondary to osteochondrosis** was diagnosed. Osteochondrosis is most often described in young, rapidly growing bulls, and particular lineages are predisposed.

Two 6-month-old calves out of a mob of 180 animals in the Manawatu were found recumbent, with nystagmus, blindness and bruxism. They had normal temperatures and failed to improve after injection with a B1/B12 supplement. Both died. Brain tissues submitted for histology confirmed the presence of **polioencephalomalacia**, which in ruminants is caused by thiamine deficiency induced by dietary changes and altered ruminal flora, which causes excessive thiaminase activity. Alternatively, excessive dietary sulphur may cause the production of toxic hydrogen sulphide gas, which interferes with cellular metabolism in the brain, also resulting in polioencephalomalacia.

Four dairy cows from a herd near Carterton died over a period of one month. They all had similar signs: decreased production with bloating, rumen stasis and death within 48 hours. The cows were on a grass-only diet. Necropsy revealed numerous white nodules on the serosal surface of the large intestine. Histologically, these corresponded to the presence of multifocal transmural eosinophilic microabscesses affecting the colon submucosa, muscularis and serosa. Migrating nematodes of the genus **Oesophagostomum** were considered to be the most likely cause of the lesion. *O. radiatum* can cause significant clinical disease, more typically in younger stock,

with diarrhoea, hypoproteinaemia, anaemia and sudden death.

A group of 10 calves in Taupo were well managed and drenched very regularly. Two died suddenly after a short disease course involving diarrhoea, dehydration and loss of condition. Culture of faeces revealed the presence of **Salmonella Typhimurium phage type 23**, which was likely the main cause of disease in this case.

Ten out of 300 heifers on a Taupo property died suddenly. There were few abnormalities visible at gross postmortem. However, a large quantity of sheep faeces had been dumped on the paddock before the deaths, followed by 100–150 mm of rain, creating the potential for large amounts of nitrogen to be washed into the soil. The aqueous humour nitrate level in one animal was > 25 mg/L (toxic level > 25), indicating that **nitrate/nitrite toxicity** was the most likely cause of death. Actively growing plants, particularly following a period of drought or dry weather in well-fertilised soils, tend to have the highest risk of causing nitrate toxicity.

Three dairy calves from a mob in the Horowhenua district died with evidence of lice, poor body condition and respiratory distress. They had a moderate to marked cranioventral pneumonia with consolidation of lung lobes visible grossly, accompanied by marked fibrinous pleuritis. Histologically there was a marked acute severe fibrinous and necrotising bronchopneumonia. Culture of the affected lung yielded **Bibersteinia trehalosi** (formerly known as *Pasteurella trehalosi*). While this pathogen has been associated with pneumonia deaths in calves, recent work involving tracheal inoculation of calves suggests that it is unlikely to behave as a primary pathogen (Hanthorn *et al.*, 2014).

A 6-year-old Ayrshire dairy cow in the Kaipara district collapsed suddenly and died. A field necropsy performed by the submitting veterinarian revealed extensive haemorrhage into the thoracic cavity and peritoneal sac. The cow appeared grossly pale and anaemic. Examination of the submitted heart and proximal aorta revealed a 10 mm tear in the wall of the aorta above the lateral cusp of the aortic valve, which was the likely source of the haemorrhage. The liver copper level was 66 $\mu\text{mol/kg}$

(reference range 95–2 000), suggesting that this animal had **ongoing copper deficiency**. Copper is a key component of lysyl oxidase, an enzyme responsible for collagen and elastin crosslinking. Failure of crosslinking caused by copper deficiency results in weakness of arterial walls and rupture, followed by acute haemorrhage and death, as in this case.

A group of autumn-born calves 10–21 days old were purchased from a saleyard in the Waikato. About a week later 12 of them were lethargic, pyrexia, anorexic and scouring. Testing for rotavirus, coronavirus and cryptosporidia was negative, but two of them were positive for **Salmonella Typhimurium phage type 9**.

In March, two 7-month-old calves from the Matamata-Piako district were lethargic and anorexic. They had been drenched previously with ivermectin. An oak tree was present in the paddock where the calves were grazing. Biochemistry on both animals revealed marked elevations in urea and creatinine, consistent with acute renal failure. Based on the biochemistry, clinical findings, and the numerous acorns seen in the paddock, a diagnosis of **oak bud toxicity** was made. The toxic principle in acorns and oak leaves is considered to be tannins and metabolites of tannins, which cause acute renal tubular necrosis.

Facial eczema has been a problem in grazing ruminants this year, but perhaps not as severe as in other years because drought conditions have not been widespread and the weather has been cooler. In a typical case, a mature cow was seen down with evidence of irritation of the udder and an elevated temperature. Moderate to marked elevations were seen in AST (477 IU/L; reference range 0–179), GLDH (494 IU/L; reference range 8–41) and GGT (660 IU/L; reference range 0–36). While haemolytic anaemia is sometimes associated with sporodesmin ingestion, there were no haematologic abnormalities in this particular animal.

EQUINE

A Stationbred gelding on the Kapiti coast developed a large mass of granulation tissue at the castration site, though it was three years since this procedure had been performed. The horse appeared bright and had a normal temperature. Histology of tissues taken from this site revealed a

severe pyogranulomatous inflammation with fibrosis, accompanied by numerous small colonies of cocci surrounded by proteinaceous material. A diagnosis of **chronic staphylococcal infection**, likely caused by *Staphylococcus aureus*, was made. It was suspected that the infection may have been present since castration.

A Thoroughbred foal about 6 months old from the Auckland region had intractable diarrhoea, with severe hypoalbuminaemia and fever. Faeces were submitted for PCR testing for *Salmonella* spp, equine rotavirus, *Lawsonia* spp, enterotoxigenic *Clostridium perfringens*, *Rhodococcus*, *Cryptosporidium*, toxigenic *Clostridium difficile* and equine coronavirus. *Lawsonia* DNA was detected, suggesting that **proliferative enteropathy** caused by *Lawsonia* may have been the underlying cause of the diarrhoea. Equine proliferative enteropathy is frequently diagnosed from clinical findings and PCR results. Since it typically responds well to treatment, cases rarely reach the stage of post-mortem examination.

A 14-year-old Clydesdale mare had multiple scabs and hair loss on the side of its face, muzzle and eyes and the signs appeared to be spreading. Routine cytology on skin scrapes revealed no significant abnormalities, but a KOH digest on hairs and keratin from the affected area revealed numerous fungal arthrospores. Dermatophyte culture revealed the presence of *Trichophyton equinum*, a common cause of **dermatophytosis** in horses.

AVIAN

An 8-year-old sun conure (*Aratinga solstitialis*) in a collection in the Waikato was treated with moxidectin for a *Capillaria* infection, but died 3 days later. On necropsy the bird was found to be emaciated. Histologic examination confirmed the presence of numerous worms in the intestine, with biopercolate eggs consistent with a group of parasites identified as *Capillaria* spp. However, the nomenclature of this group of parasites is disputed, with some taxonomists classifying them in a single genus (*Capillaria*) and others dividing them into 22 different genera. The large number of potential genera for this group of parasites reflects the diversity of hosts and life cycles. Some of the parasites have direct life cycles, while others require an intermediate host. In

addition, some of the parasites in this group show evidence of anthelmintic resistance. In this particular case, all of the worms remaining in the carcass appeared viable, suggesting that the anthelmintic treatment may have been ineffective. Resistance to moxidectin in some species of *Capillaria* is considered to be relatively common.

CANINE

A dog at a pound in Northland died acutely. On necropsy there was evidence of dehydration. The small intestine appeared reddened and there was fluid in the stomach, with dark brown sludge in the small intestine. On histology the small intestine was found to be autolysed, but there were regions of extensive mucosal necrosis visible in some areas, with thrombosis of mucosal blood vessels. There were numerous long bacilli resembling *Clostridium* invading the necrotic area. Histologic lesions were considered consistent with **acute clostridial enteritis**, which in dogs has been attributed to the toxin-producing species *C. perfringens* and *C. difficile*. While many dogs may subclinically carry small numbers of bacteria, a change in the enteric environment favouring the proliferation of clostridial species may precipitate rapid bacterial proliferation and toxin production. Assays to demonstrate toxin in ingesta are not routinely available for *C. perfringens* but they are available for *C. difficile* as this agent is also an important cause of antibiotic-related enterocolitis in humans.

A two-month-old puppy in the Waikato died suddenly a week after routine vaccination. A necropsy was performed by the submitting veterinarian, who noted gross abnormalities in the heart, which was submitted to the laboratory for histologic examination. On gross examination by the pathologist, the heart appeared globose and had pale areas in the muscle. These pale areas corresponded histologically to large regions of necrosis, which were accompanied by a thick layer of neutrophils at the margins of the necrosis and extended into the surrounding, more normal tissue. The **marked necrotising and suppurative myocarditis** was considered to reflect a bacteraemia, possibly the result of an umbilical infection. Microbiology was not performed.

A five-month-old Poodle cross from the Auckland region had soft faeces for 12 days prior to presentation to the local veterinarian. The dog had been fed a raw-food diet from the age of 10 weeks. Cultures for *Salmonella* and *Campylobacter* were negative and there was no evidence of intestinal worms, but *Giardia* was detected by antigen ELISA. A diagnosis of **giardiasis** was made. *Giardia* is not typically associated with raw-food diets, but is more frequently a water-borne pathogen.

PORCINE

Twenty weaner pigs were brought into a small piggery on a dairy farm near Carterton. Four of them died over a period of about 24 hours. The pigs had a bluish discolouration of the skin on the ventral abdomen prior to death. Post-mortem examination revealed haemorrhage along the greater curvature of the stomach and intestines, histology demonstrated the presence of a necrosuppurative enterocolitis, and there was multifocal necrosis in the liver. A **bacterial enteritis** with extension to **bacteraemia/septicaemia** was considered the most likely cause of these lesions. A **K88 positive *Escherichia coli*** was isolated from the intestinal contents, but generally enterotoxigenic *E. coli* species like this do not produce such severe signs, so it was unlikely to have caused the lesions and sepsis seen. Sepsis caused by another *E. coli* type, or by other enterobacteria such as *Salmonella*, remains the more likely cause of death in this case.

LAGOMORPH

Six rabbits and two guinea pigs belonging to a private owner died over a 3-week period, with no premonitory clinical signs. Necropsies were performed by the submitting veterinarian and no gross abnormalities were noted. Histology revealed random periportal to midzonal hepatocellular necrosis and disassociation in the liver. Glomeruli in the kidney and the alveolar blood vessels of the lung contained multiple small fibrin thrombi. These findings were diagnostic for **rabbit haemorrhagic disease** caused by **rabbit calicivirus**. This virus only affects rabbits, so the cause of deaths in the guinea pigs in this case remained unknown.

Gribbles Veterinary Pathology

BOVINE

A Taranaki dairy farmer reported an outbreak of acute illness and death in five-month-old crossbred Friesian replacement heifer calves. Affected animals presented with respiratory distress and dehydration prior to recumbency and death. Seven calves in the group of 130 animals died over a period of five days. Gross post-mortem findings included fibrinous peritonitis and pleuritis. Histopathology confirmed the presence of fibrinous polyserositis accompanied by numerous small bacterial rods. *Pasteurella multocida* was cultured from the lung. The findings were considered consistent with **septicaemic pasteurellosis** caused by *Pasteurella multocida* capsular type B.

Four out of 70 five-month-old Friesian heifer calves on a Franklin district farm were observed to be isolated, uncomfortable and hunched up, and one became recumbent and died. The mob had been yarded the previous day and treated with pour-on anthelmintic and subcutaneous injections of vitamin B12, selenium and copper at the appropriate dose rates. Post-mortem examination of the dead calf was non-diagnostic. Samples of liver, kidney, heart and rumen were collected for histopathology. The liver was disrupted by diffuse hepatocellular necrosis characterised by dissociation, hypereosinophilia and pyknotic or karyorrhectic nuclei. There was haemorrhage throughout centrilobular zones. Serum biochemistry from one of the sick calves revealed a marked increase in GLDH (1 460 IU/L; reference range 5–35), increased bilirubin (16 µmol/L; reference range 0–8) and normal GGT (42 IU/L; reference range 3–47), indicating severe hepatocellular damage and possible haemolysis. In light of the history, these findings were considered compatible with **acute copper toxicity**.

Three spring-calving mixed-age Friesian cows from a South Waikato herd of 300 aborted over several days. All had grossly visible placental thickening and exudate. Dam sera were negative for *Neospora caninum* by IFAT and also for bovine viral diarrhoea virus using PCR. Histopathology revealed necrosis, inflammation and vasculitis in

the placenta, with scattered branching fungal hyphae, consistent with **mycotic abortion**. Culture of fetal stomach contents produced a light growth of *Aspergillus fumigatus*.

An adult Angus beef cow, one of 12 cattle on a small lifestyle farm in South Canterbury, was presented for veterinary examination because it was straining and had been losing weight. Rectal palpation revealed many hard masses in the perirectal tissues. The animal was euthanased and samples of the mass submitted consisted of necrotic fat. Clinical and histological findings were consistent with the animal having the condition known as **abdominal fat necrosis**. This condition is believed to develop in cattle grazing tall fescue pasture, but this farm had little of this grass species.

Three rising-two-year-old Friesian heifers in a mob of 120 on a South Canterbury dairy farm died over a two-day period and one was necropsied. The carcass was distended with gas, and froth was coming out of the nose. Histological examination revealed an embolic septic process in the lung with large numbers of slender, Gram-negative rods consistent with *Fusobacterium* sp. in vessels. Anaerobic culture of the lung obtained a pure growth of a *Fusobacterium* sp., resulting in diagnosis of an **embolic fusobacterial pneumonia**. This bacterium is present in the alimentary tract and may gain access to the circulation from anywhere along the alimentary tract, especially when the wall of the rumen is compromised by severe inflammation from carbohydrate overload. The rumen in this case was normal and the source of the bacteria was not determined.

A single rising-two-year-old Friesian heifer in a group of 40 on a Canterbury farm had several lumps at the angle of the jaw. Biopsy of one of these lesions revealed typical lesions of **actinobacillosis** caused by *Actinobacillus lignieresii*.

A coastal farm near Motunau in North Canterbury had a group of 60 Jersey bull calves grazing pasture and receiving supplemental balage. Three calves became sick and one died. The animals had clinical evidence of photosensitivity, with oedema of the brisket in one and oedema of the scrotum in all three. Feed was in short supply and when

grazing a paddock along the driveway the calves had eaten leaves of trees growing through the fence. These trees were identified as ngaio (*Myoporum laetum*) and a diagnosis of **ngaio toxicity** was supported by finding leaves of this species in the rumen of the dead calf. Ngaio is a mainly coastal tree that causes a hepatogenous photosensitivity, and biochemical evidence of liver damage was found in the two live calves. Levels of GLDH were 309 and 104 (reference range 5–35), levels of GGT were 171 and 67 (reference range 3–47) and bilirubin levels were 28.0 and 12.8 (reference range 0–8).

A calf died suddenly on a North Canterbury farm. It was the seventh death in a mob of 150 calves over a period of 5–6 weeks. At postmortem the calf had a diffuse fibrin-rich peritoneal exudate. Histological examination confirmed an acute peritonitis, and culture of the exudate yielded a pure growth of *Pasteurella multocida*. Examination at the Animal Health Laboratory (AHL), Wallaceville confirmed that the isolate was type B and consistent with previous isolates from cases of **systemic pasteurellosis** seen in calves of this age.

Fresh and fixed lung was received from a dairy cow that had died suddenly after a 24-hour illness. The cow was housed in an open-sided barn in South Canterbury. It was the fourth cow in a week in a herd of 1 200 to acutely develop the same problem. Histologically the cow had an **acute fibrinous pneumonia**, and *Mannheimia haemolytica* was recovered in pure culture. This organism is a common cause of acute fibrinous pneumonia in goats and sheep in New Zealand, but an uncommon cause of pneumonia in cattle.

A 17-month-old Friesian heifer from a herd of about 150 in North Canterbury had generalised proliferative dermatitis covering most of the dorsal trunk. It was most severe around the head and neck. She was otherwise bright and had a normal temperature. While having recently lost some weight, she was not in noticeably poorer body condition than the rest of the group. A sample of scabs and hair from this animal stained with methylene blue showed numerous organisms typical of *Dermatophilus congolensis*, the causative organism of **dermatophilosis**. Also seen were fungal

spores and hyphae, suggesting that this animal had concurrent **dermatophytosis** (ringworm). The species of dermatophyte was not identified as a fungal culture was not requested. The presence of two concurrent infections raised questions about the immunocompetence of the heifer, but a test for bovine viral diarrhoea virus antigen was negative and blood selenium was within normal range.

In a mob of 40 well-grown calves on a Southland dairy farm, seven died over a short period after 2–3 days of dyspnoea and dullness. A necropsy of one dead calf showed severely consolidated lungs and yellow fluid in the abdomen. Histology revealed a severe acute bronchopneumonia and *Histophilus somni* was detected in the fixed lung by PCR. Culture was attempted initially but the lung sample was heavily contaminated.

In another case, on a dairy farm in the same area of Southland and during the same week, eight calves in a mob of 300 died over a 5-day period. It was raining and cold at the time. Necropsy of two of the dead calves showed abundant fibrinous effusion in the chest and abdomen of both. A heavy, pure growth of *Pasteurella multocida* was isolated from the effusion from one calf, consistent with a diagnosis of **systemic pasteurellosis**. The remaining calves were treated with a long-acting tetracycline antibiotic and there were no further cases.

A very sick cow with severe, watery diarrhoea was examined. It was one of a Southland dairy herd of 800 cows. A heavy, pure growth of *Salmonella Typhimurium* was isolated from the faeces. Another cow had died a few days earlier with similar signs.

Three of 50 four-month-old calves on a Southland property were found recumbent with severe diarrhoea, dehydration and pyrexia. A heavy growth of *Yersinia pseudotuberculosis* was cultured from the faeces of all three. There were 12 other similar outbreaks of yersiniosis throughout Otago and Southland during the reporting period, mostly in January. Affected calves were reported to be ill-thrifty, with diarrhoea.

On a Southland beef farm twelve 18-month-old beef bulls in a mob of 60 died over a 1-month period, usually about a week after the onset of severe diarrhoea and wasting. Necropsy of a

recently dead bull showed no significant gross lesions but histology revealed lesions in the ileum consistent with acute **mucosal disease**. This diagnosis was confirmed by a positive **bovine viral diarrhoea (BVD)** antigen ELISA test on a blood sample taken at necropsy. All remaining animals were bled and tested for BVD by pooled PCR and another 10 persistently infected bulls were identified.

Two 2-year-old beef bulls in a mob of 90 were found dead on a large Otago sheep-and-beef farm and several others looked unwell. Necropsy of one dead bull revealed blood in the pleural cavity and petechial haemorrhages over the heart. Histology of sections of the heart revealed large areas of chronic active myonecrosis consistent with **monensin toxicity**. Further investigation found that a proprietary product containing monensin had been recently added to the water supply and after this was removed no more deaths occurred.

Ten deaths occurred over a 2-week period in a group of 140 well-grown six-month-old calves on a Southland dairy farm. Affected animals were found dead or were noticed to be unwell and recumbent several hours before death. Necropsy of one recently dead calf was unremarkable apart from a small amount of straw-coloured, fibrinous fluid in the abdomen. Histology on a range of fixed tissues revealed a severe bacterial peritonitis. Unfortunately no fresh samples were submitted to the laboratory so culture was not possible, but from the history and clinical and postmortem findings a *Pasteurella multocida* infection (**systemic pasteurellosis**) was suspected. The whole group was injected with a long-acting tetracycline antibiotic and there were no more deaths.

Twelve cows from a 500-cow dairy herd in Southland were affected by a severe upper respiratory tract infection. The clinical signs included a purulent nasal and often ocular discharge, occasional coughing, anorexia and a marked drop in milk yield. These cows were not pyrexial and did not respond to antibiotics. **Infectious bovine rhinotracheitis (IBR)** was suspected and samples of nasal discharge tested positive for IBR by PCR.

Two dairy calves from a mob of 20 in Northland died suddenly. There were no abnormalities identified at postmortem and a range of fixed tissues were submitted for histopathology. Tissues

were variably autolysed but in the small intestine it was possible to recognise scattered colonies of coccobacilli associated with necrosis and neutrophil infiltrates within Peyer's patches. In the kidney, there were scattered amphophilic intranuclear inclusion bodies within endothelial cells. These findings were consistent with **combined yersiniosis and infection by bovine adenovirus**. Bovine adenovirus is often associated with acute enteric disease and death in young cattle, and may have been the primary pathogen in these calves.

OVINE

Three ewes from a flock of 1 500 on a Waitomo district farm became blind, depressed and recumbent after yarding and shearing. Two were euthanased and various tissues were submitted for histopathology. The brains of both had symmetrical vacuolation of grey-and-white-matter junctions and Alzheimer type II astrocytosis. Hepatocytes contained clear lipid vacuoles. Liver vitamin B12 concentrations were 110 and 101 nmol/kg (adequate range 200–1 500). The ewes were diagnosed with **hypovitaminosis B12, hepatic lipidosis and hepatic encephalopathy**.

About 20 lambs from a Hastings district farm developed apparent blindness and neurological signs while grazing a recently irrigated sorghum crop. Histological evaluation of a brain from an affected lamb revealed numerous spheroids scattered throughout the vestibular nuclear complex, cerebellar nuclei, and midbrain nuclei accompanied by white-matter vacuolation and astrogliosis. Neuraxonal spheroids develop as a result of intrinsic (metabolic) or extrinsic (toxic) disturbance of the function of axons or as a result of chronic neuronal injury. Causes include vitamin E deficiency, various plant toxicities, storage diseases, hereditary neuraxonal dystrophies and experimental toxicities. Notably, sorghum toxicity is reported to cause lesions of neuraxonal degeneration in sheep (Bradley *et al.*, 1995). The toxic agent is unknown but thought to be related to that causing lathyrism in humans and animals. Given the histological lesions and circumstantial evidence, **sorghum toxicity** was considered the most likely diagnosis in this case.

Clinically significant **coccidiosis** is not commonly diagnosed in laboratory

submissions from sheep, but was suspected in an outbreak of diarrhoea and death in lambs on a Rangitikei district farm. About 200 lambs in a mob of 300 developed diarrhoea and more than 15 died. An appropriate worm-drenching programme was in place and nematode faecal egg counts were not particularly high (up to 250 per gram). Post-mortem examination and histopathology conducted on an affected lamb showed lesions that were largely restricted to the colon. There was diffuse mucosal atrophy, multifocal erosion, and widespread crypt dilation with enterocyte necrosis and regeneration. Moderate numbers of coccidial oocysts were identified within crypts. Clinical ovine coccidiosis can occur in heavily stocked pasture where the oocyst challenge is high and where lambs are relatively naive prior to exposure.

A group of thirteen 5-month-old ewe lambs in North Canterbury were treated with a triple combination anthelmintic drench (abamectin, oxfendazole and levamisole) that also contained selenium and a dietary supplement. The supplement included vitamins A, D and E; magnesium, sodium and calcium; trace elements (copper, zinc, iron, iodine, boron, cobalt and selenium); seaweed, carbohydrates, amino acids and a herbal extract. Six lambs died over the next three days and three more were unwell when a veterinarian was called to the property. On checking the dose rate it was found that the animals, whose body weight was around 30 kg, had been given the combination drench at the recommended dose for a 50-kg animal. Liver collected from one of the dead animals had a selenium concentration of 28 530 nmol/kg (toxic range >3 000), which was suspicious for **selenium toxicity**.

On a small Otago sheep farm with a mixed-age mob of 275 ewes (hoggets and older ewes), six ewes were found dead or ataxic and stargazing shortly before becoming recumbent and dying. Histology of the fixed brain from one of these dead ewes showed lesions typical of **polioencephalomalacia**.

In a small mob of 15 mature rams on an Otago sheep farm, two rams died and one was off-colour with severe diarrhoea. Acute salmonellosis was suspected but culture of faeces from the sick ram was negative for *Salmonella* spp.

Instead a heavy growth of *Yersinia pseudotuberculosis* was isolated. This is an unusual isolate from an adult ruminant: most outbreaks of yersiniosis occur within the first year of life.

On a large Central Otago sheep farm at least 20 of a mob of 150 well-grown two-tooth rams were found to have large goitres. One ram was euthanased for necropsy and one of its thyroid lobes measured 120 mm in length and 60 mm in width (normal 20–30 mm). Histopathological examination of the thyroid confirmed a severe hyperplasia consistent with an **iodine deficiency**. Serum samples from 11 of the affected rams contained only 3–13 µg/L of iodine (deficient level < 20), consistent with exposure to an iodine-deficient pasture. They had been wintered on swedes and then fed a mix of pasture and chicory.

EQUINE

On separate farms in Canterbury, two cases of **fungus endometritis** were diagnosed in mares with a history of failure to conceive. One mare was described as having a pool of fluid in the uterus. At the laboratory the fluid was noted to be white and cloudy, and cytological examination showed it to be highly cellular, with degenerate inflammatory cells (including some eosinophils) and large numbers of yeasts and pseudohyphae. Identification by MALDI-TOF revealed the presence of *Candida guilliermondii*. In the second case, the mare also presented with a large amount of fluid in the uterus and there was no history of prior antibiotic administration. Fluid was greyish white and cloudy, and cytological examination revealed large numbers of degenerate neutrophils and rare eosinophils. Bacteria and yeasts were also found. Microbial culture revealed large numbers of *Enterobacter aerogenes* and large numbers of a *Candida* sp. that was not *C. albicans*.

CAPRINE

Skin biopsy samples were submitted for evaluation from three Saanen cross goats on a Waikato dairy farm. About 20 of 600 goats on the farm had developed proliferative skin lesions on the mouth, feet, legs and teats over the preceding few weeks. Microscopic examination of the skin revealed severe ulcerative and crusting dermatitis with areas of ballooning degeneration of the epidermis. Some of the ballooned cells contained globular amphophilic

structures consistent with poxviral inclusion bodies. These findings confirmed the clinical suspicion of **parapoxviral dermatitis (scabby mouth)**.

FELINE

A 9-year-old male Siamese cat from New Plymouth had a swollen hind foot and toe with a discharging sinus. A week of treatment with amoxicillin/clavulanic acid at recommended dose rates produced no improvement. An aspirate of material from the region showed pyogranulomatous inflammation. There were very low numbers of filamentous bacteria present and a Gram stain revealed filamentous Gram-positive rods in very low numbers. There was a light growth of *Nocardia* sp., which on sensitivity testing was found to be resistant to amoxiclav, cephalothin, enrofloxacin, penicillin, tetracycline, trimethoprim/sulpha and ampicillin, but sensitive to erythromycin. It was therefore infection with a **multi-drug-resistant *Nocardia* sp.**

In a litter of six 11-week-old domestic shorthair cats from Auckland, one died and two became unwell. One recovered with supportive care but the other deteriorated and was euthanased. A range of tissues were collected at postmortem and submitted for histopathology. A sample of lung showed multifocal broncho-interstitial or embolic pneumonia with intralesional Gram-negative bacilli. A sample of small intestine contained neutrophilic enteritis with villus atrophy and regeneration, and protozoa in crypts, alongside villi or in the lumen. These sometimes appeared to have one or two nuclei. A combination of pneumonia caused by *Bordetella bronchiseptica* and enteritis caused by a **mixed infection of *Trichostrongylus axei* and *Giardia* sp.** was suspected. This was supported by a follow-up faecal sample from the dam, which was positive for *T. foetus* by PCR and positive for giardia by antigen ELISA.

CANINE

A two-year-old male Heading dog progressively developed severe respiratory distress, mucus membrane cyanosis and diarrhoea over a period of five days. The clinical signs reportedly developed after neighbouring paddocks were sprayed with herbicide, including the possible use of paraquat. The dog was euthanased and samples collected for histology. Significant lesions were

confined to the lungs, where there was diffuse type II pneumocyte hyperplasia, fibrin exudation into airways, alveolar emphysema, and interstitial fibrosis and haemorrhage. This pattern of interstitial pneumonia was considered compatible with **paraquat toxicity**. Paraquat is an organic viologen used as a non-selective herbicide. It is exquisitely toxic to mammals when ingested (LD50 22–262 mg/kg). Large doses cause acute pulmonary oedema, renal necrosis and death, while smaller doses cause fatal interstitial pneumonia in 5–10 days. The toxin is absorbed from the GI tract, then accumulates in and targets the delicate type I alveolar pneumocytes, resulting in their loss and replacement by cuboidal type II pneumocytes.

An 8-year-old dog of unspecified breed from a farm in the Whanganui district was anorexic, lethargic and showed some abdominal discomfort. There was no vomiting or diarrhoea and it was unclear whether polydipsia or polyuria was present. Routine biochemistry revealed azotaemia. Creatinine was 192 µmol/L (reference range 42–109) and urea was 44.2 mmol/L (reference range 2.5–9). Liver enzymes and other biochemical parameters and haematology were normal. However, serology revealed a *Leptospira interrogans* Pomona titre > 1:1 600, a Hardjo titre of 1:100 and a Copenhageni titre of 1:200, consistent with *Leptospira interrogans serovar Pomona* infection.

An 11-year-old spayed Border Collie in the Whanganui district presented with a large intraluminal bladder mass at the cranial pole. An ultrasound-guided fine-needle aspiration was carried out. Clusters of epithelial cells, some of which were morphologically uniform with mild anisocytosis and anisokaryosis, were seen in a background of blood. Cells had round nuclei, finely stippled chromatin and inconspicuous nucleoli. There were occasional clusters of cells showing a marked variability in the nucleus-to-cytoplasm ratio. Some clusters contained cells with coarse chromatin patterns and these cells were often very large with large, prominent nucleoli. These findings supported a diagnosis of **transitional (urothelial) cell carcinoma**.

An 8-year-old female dog of unknown breed was presented to a Taranaki veterinary clinic. She was sick and inappetent, and although she did improve, she was straining and unable to

urinate. Clinical examination revealed a huge bladder and no neurological deficits. An exploratory laparotomy revealed an enlarged nodular neck of bladder, which was sampled by fine-needle aspiration. The majority of cells present were well-preserved epithelial cells showing marked anisocytosis and anisokaryosis, and some were very large. Cells showed a variable nucleus-to-cytoplasm ratio. The nucleus tended to be round with coarse chromatin patterns and 1–4 large nucleoli that were occasionally of greater diameter than a red blood cell. There were occasional binucleated cells. This was a carcinoma, likely to be a **transitional (urothelial) cell carcinoma**.

A three-month-old Cavoodle from Auckland had vomiting, diarrhoea, lethargy and depression for three days. A faecal sample yielded moderate to heavy numbers of *Isospora* sp. oocysts, suggesting that the puppy had **coccidiosis**.

A nine-month-old Bull Terrier from Auckland had a nasal discharge for one month. Computer tomography scanning and rhinoscopy showed areas suspicious for osteomyelitis or fungal infection. Histopathology of nasal biopsies showed neutrophilic and lymphoplasmacytic rhinitis, along with mats of branching fungal hyphae and fan-shaped conidia on conidiophores, consistent with **nasal aspergillosis**. This was supported by culture of *Aspergillus fumigatus* from a nasal tissue sample.

CAMELID

A 13-day-old alpaca cria from the Auckland region had bilateral effusion and pain in multiple joints. *Klebsiella pneumoniae* was cultured from one joint aspirate, suggesting that the cria had **septic arthritis**. As in other species, this is often associated with failure of passive transfer (Dolente *et al.*, 2007).

AVIAN

There were two cases of suspected **lead toxicity** in native birds during this period. In the first case a one-year-old New Zealand falcon (*Falco novaeseelandiae*) from the Marlborough region was off its food and radiographs showed possible lead pellets in the digestive tract. Blood lead concentration was 0.04 mg/L. No toxic range has been determined for this species, but the presence of a measurable (therefore potentially significant) level in a bird with

clinical illness and evidence of exposure was considered suspicious. In the second case, a four-month-old kea (*Nestor notabilis*) from the Arthur's Pass area in the Southern Alps had been vomiting for three days and lead poisoning was suspected. Blood lead concentration was 0.7 mg/L. Again, no toxic range has been identified for this species but this result supported the clinical suspicion of **lead toxicity**.

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Quarterly report of investigations of suspected exotic diseases

Exotic vesicular diseases ruled out

A veterinarian notified MPI via the exotic pest and disease hotline of a steer with several small circular tongue lesions.

A bull on the property had had similar lesions and condition loss 3 weeks previously, had been treated for woody tongue and had recovered. There was no history of travel or animal movements onto the farm, or of overseas visitors or returnees to the farm. Photographs were examined by the Incursion Investigator and the multi-focal lesions looked granulomatous (**Figure 1**). Vesicular disease was stood down on clinical and epidemiological grounds. However, the lesions did not appear typical for an endemic disease, so testing was done at the MPI Animal Health Laboratory (AHL), Wallaceville, to confirm the cause. Swabs of the lesions tested negative for poxvirus and parapoxvirus by PCR. No *Actinobacillus* spp. were isolated on specific *Actinobacillus* culture, and general culture yielded a heavy mixed growth with no predominant isolates. Cytology from scrapings of the lesions supported a tentative diagnosis of woody tongue (*Actinobacillus lignieresii*). Histopathological examination of biopsies of the lesions confirmed pyogranulomatous glossitis. The sulphur granules present, pyogranulomatous inflammation and Gram-negative rods



Figure 1: Gross tongue lesions

Exotic disease investigations are managed and reported by MPI Diagnostic and Surveillance Services, Wallaceville. The following is a summary of investigations of suspected exotic disease during the period from January to March 2017.

within the granules were classical for woody tongue caused by *Actinobacillus lignieresii* (**Figures 2 and 3**). The unusual presentation of small, discrete lesions only on the tongue may have been caused by traumatic abrasions allowing the introduction of the bacteria, and failure to isolate the organisms by culture may

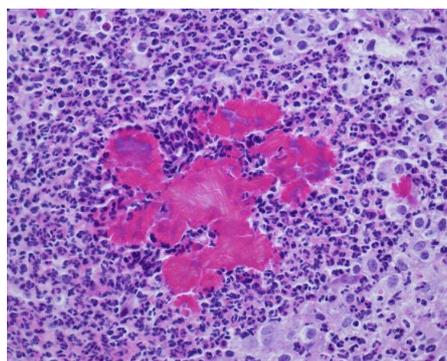


Figure 2: Photomicrograph of pyogranulomatous inflammation within the tongue of a cow, showing bacterial "sulphur granules" composed of bacteria (purple) and material thought to be produced by the host's immune reaction (magenta). All this is surrounded by neutrophils, with macrophages at the periphery. H&E stain. (Photo: Keren Dittmer)

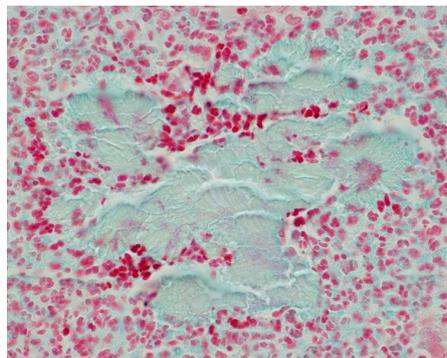


Figure 3: Photomicrograph of Gram-negative rod bacteria within "sulphur granules" in the parenchyma of a cow's tongue. Gram stain. (Photo: Keren Dittmer)

have been due to treatments given to the animal. As an endemic condition was confirmed, the investigation was stood down.

A veterinary pathologist contacted MPI via the exotic pest and disease hotline to discuss a clinical presentation in 80 of 160 steers aged 15 months. There were ocular and muzzle lesions and two animals had oral lesions and lameness that could be consistent with exotic vesicular disease. Following consultation with the farm veterinarian the duty Incursion Investigator had an AsureQuality Initial Investigating Veterinarian (IIV) visit the affected property with the farm veterinarian. The IIV confirmed that the oral lesions and lameness were confined to just two animals out of a total of 1 000 cattle on the property. Vesicular disease was ruled out on clinical and epidemiological grounds and the investigation was stood down.

Exotic infectious bovine rhinotracheitis excluded

A private veterinarian notified MPI of a herd of rising-two-year-old dairy heifers with apparent reproductive failure. Of 290 Friesian heifers mated through a synchrony programme, 130 (44 percent) were considered not to be pregnant at the time of testing. The veterinarian had observed that there were small papules on the vulva and described small vesicles in the vagina of affected heifers. These lesions were present in all non-pregnant heifers, but not in all of the pregnant heifers. However, after reviewing the mating records it was determined that the low apparent pregnancy rate was a reflection of the period that cattle had been mated prior to being tested. Follow-up blood testing of a sample of 26 supposedly non-pregnant heifers using

the pregnancy-associated glycoprotein ELISA test revealed that 17 (65 percent) were in fact pregnant. Thus reproductive performance at the time of notification was within normal limits.

Investigation carried out in parallel focused on excluding strains of bovine herpesvirus-1 (BoHV-1) responsible for infectious bovine rhinotracheitis (IBR) that could potentially cause reproductive failure. A number of presumed non-pregnant heifers (based on testing by ultrasound while heifers were still being run with bulls) and pregnant heifers were blood-tested for IBR using both VNT and ELISA. At the time of the report all heifers were negative for IBR, indicating that BoHV-1 was unlikely to be the cause of any reproductive failure (if in fact reproductive failure was present). Follow-up testing one month later showed that the seroconversion of heifers had occurred and was approximately in the same proportion for both groups (6/12 vs. 5/12; $p = 1$). This indicated that exposure to BoHV-1 had occurred recently and was likely to have been responsible for the lesions seen, but not any reproductive loss. Histological examination of biopsies provided supportive evidence leading to a diagnosis of infectious pustular vulvovaginitis (IPVV) caused by BoHV-1, but this was not shown to be the cause of reproductive failure. The investigation was stood down.

Enzootic bovine leukosis excluded

An AsureQuality veterinarian notified MPI of two 18-month-old bulls that were positive by pre-export screening ELISA for antibodies to enzootic bovine leukosis. The bulls were from a shipment of about 1 400 cattle for export to China. Samples were sent for confirmatory ELISA testing at the Livestock Improvement Company and both tested negative. The initial test results were considered to be false positives. Exotic disease was excluded and the investigation was stood down.

Haemorrhagic septicaemia excluded

A veterinarian contacted MPI via the exotic pest and disease hotline to report mortalities over a two-week period in a herd of about 700 yearling deer. The deer had been brought together from three sources about three months earlier

and were being intensively managed under a break-feeding regime on a kale crop. Two of the 28 deer that died had undergone postmortem. Gross changes indicated septicaemia and were characterised by petechiation of the mucosal and internal serosal membranes, myocardium and subcutis. In both animals the meninges were congested, with fibrinopurulent material present, and in one a frontal sinusitis was evident. Histopathology identified suppurative meningoencephalitis, and a heavy growth of *Pasteurella multocida* was cultured from one animal. This presentation was consistent with a potential diagnosis of haemorrhagic septicaemia (*P. multocida* serotypes B:2 and E:2), so the culture was submitted to the AHL (Wallaceville) for further assessment. Molecular assays excluded haemorrhagic septicaemia serotypes. The presentation had similarities to multiple outbreaks of septicaemic pasteurellosis reported in calves in New Zealand, caused by *P. multocida* capsular type B (McFadden *et al.*, 2011). *P. multocida* is considered part of the normal flora of the upper respiratory tract in a wide range of animals. Both septicaemia and pneumonic forms of pasteurellosis are recognised in deer, apparently precipitated by a number of stressors including transport, overcrowding, inclement weather and high parasite burdens (Mackintosh *et al.*, 2002; Wilson, 2002). Multilocus sequence typing is being carried out to determine the capsular type and strain involved here. The mob was moved on to a grass pasture and no further deaths occurred. Exotic disease was excluded and the investigation was stood down.

Chlamydophila abortus excluded

A veterinary pathologist phoned MPI to report an outbreak of sheep abortion, ewe deaths and pinkeye among sheep in North Canterbury. Earlier in the same season, mixed-age ewes in the flock had experienced conjunctivitis in conjunction with and followed by abortions, starting in August and continuing into September. Placentas were reported as being “thickened and rubbery” but no histology was performed. By the time of notification, the property had progressed from ewe abortions and was experiencing hogget abortions and death of neonatal lambs. Very young lambs

from hoggets (as young as 2 days old) had been reported with conjunctivitis. About 75 lambs produced by the mob of 340 hoggets had died, but the total number of live lambs was not known. In addition, up to 30 ewes out of about 650 had died. Swabs were collected from all types of lesions, including conjunctivitis lesions, and vaginal swabs were taken from ewes and hoggets. No single cause of the signs was found. The agent of the exotic disease ovine enzootic abortion, *Chlamydophila (Chlamydia) abortus*, was ruled out by generic *Chlamydia* PCR, followed by sequencing of a single positive sample, which was identified as *C. pecorum*, a known cause of sporadic disease in lambs. Bacterial culture in Friis broth, followed by PCR and genetic sequencing of products, was positive for *Mycoplasma conjunctivae* in multiple conjunctivitis samples. A single placenta contained an unusual microscopic pattern of vasculitis but no apparent placentitis. Other placentas and fetuses submitted for pathological examination were non-diagnostic. The major exotic differential diagnosis in this case was excluded and a possible cause of the lamb conjunctivitis was found, but no cause of abortions or ewe deaths was found. This is likely to be due to timing and the limited availability of representative samples, with samples being collected only in October. Exotic disease was ruled out and the investigation was closed.

Hydatids excluded

An MPI Veterinary Technical Supervisor called the exotic pest and disease hotline to report finding cysts in the livers of three sheep at post-mortem inspection in a meat processing plant. The aged ewes were from a line of 317 from a trading property that purchased animals from several source farms. The specimens were sent directly to a veterinary pathologist for gross and histological examination, with instructions to send on samples to the AHL (Wallaceville) if hydatids was not ruled out. The cysts were biliary cysts of no concern and the investigation was stood down.

Caprine piroplasmiasis excluded

A veterinary pathologist called the MPI exotic pest and disease hotline to report three mildly anaemic goats. One animal had mild structural changes to the red blood cells but no evidence of

blood parasites was seen. Testing at the AHL (Wallaceville) for piroplasmosis (including *Theileria* and *Babesia* spp. organisms), anaplasmosis and haemotrophic mycoplasmas by PCR was negative. Haematology revealed that the goat with the mild erythrocyte changes had inflammation that was likely septic in origin, and the other two animals had good evidence of recurrent or ongoing endoparasitism. As exotic causes of the anaemia were excluded, the investigation was closed.

Equine herpesvirus-1 myeloencephalitis excluded

A veterinarian from Massey University notified MPI that two foals from the same farm in Palmerston North had died acutely within 3 days. The foals were 3 months old and located in the same paddock, where no toxic causes were evident and no supplementary feed was being given. The first dead foal had pulmonary oedema with acute necrosis of the lungs, pleural effusion, and some hepatic necrosis, whereas the second foal had a grossly enlarged liver with significant acute hepatic necrosis. Massey University scientists conducted bacteriology work and sent water samples from the troughs for toxin analysis, while the AHL (Wallaceville) performed virology and leptospirosis testing on samples of spleen, liver and pleural fluid from the second foal. PCR testing was negative for equine adenovirus 1 and 2 (subcontracted to Cornell University, USA), equine herpesvirus-1 and 4, equine influenza virus, equine viral arteritis and *Leptospira* spp. Virus isolation was also negative. No pathogenic organisms were found on bacteriological culture. The cause of the deaths was not established, but no further animals became ill. Since exotic causes of disease were excluded, the investigation was closed.

Alpaca piroplasms excluded

A veterinary pathologist notified MPI via the exotic pest and disease hotline of a severely anaemic 4-year-old alpaca that had died. PCR testing conducted by the AHL (Wallaceville) was negative by PCR for piroplasms including *Babesia* and *Theileria* spp., *Anaplasma phagocytophilum* and haemaphysalins (*Mycoplasma* spp.). Haematology was conducted on blood taken before the animal died, and the nature of the

anaemia and the erythrocyte morphology changes supported the likelihood of a *Haemonchus contortus* parasite burden. Other differentials could have been low vitamin B12 status, hypophosphataemia, low iron or low copper. No other animals in the herd appeared clinically unwell, but from these results the veterinarian was able to advise the owners on future management of their other alpacas.

Equine viral arteritis and equine infectious anaemia ruled out

A 6-year-old Pinto gelding with ventral and leg oedema was reported to MPI by a veterinary pathologist via the exotic pest and disease hotline. Biochemistry and haematology were consistent with equine viral arteritis (EVA) but also for several inflammatory conditions or other illnesses. The horse was stiff and sore, and while the oedema resolved over the next week, the blood changes and stiffness were still present. Two other horses on the property were unaffected. The unwell horse had only left the property once in the previous 12 months, for a show where it was in contact with other horses. Exotic causes of oedema were tested for at the AHL (Wallaceville), and the horse was negative for equine infectious anaemia, piroplasmosis and anaplasmosis. EVA testing by VNT returned inconsistent results, so this test was subcontracted to the OIE Reference Center for Equine Viral Arteritis, University of Kentucky, USA for verification and was negative. As exotic disease was excluded, the investigation was closed.

A veterinary pathologist called the exotic pest and disease hotline to report three mildly anaemic Crossbreed horses out of five at a property in Auckland. Haematology indicated that the anaemia was probably due to inflammation, but there was no clinical inflammation evident. Blood samples from two of the horses were sent to the AHL (Wallaceville), where they tested negative for exotic diseases of concern (equine viral arteritis, equine infectious anaemia, piroplasmosis and anaplasmosis), so the investigation was stood down.

A veterinary pathologist called the exotic pest and disease hotline to report a 4-year-old Thoroughbred gelding with anaemia and signs of acute inflammation. The horse had become very unwell a week earlier and had some swelling in

the submandibular region. Haematology and biochemistry were consistent with acute inflammatory changes, but no cause was identified. The horse had no history of overseas travel or contact with imported horses. Blood samples taken for exotic disease testing at the AHL (Wallaceville) tested negative for equine infectious anaemia, equine viral arteritis, piroplasms and *Anaplasma phagocytophilum* by AGID, VNT, and PCRs respectively. The horse's condition deteriorated and it was euthanased. On post-mortem examination, lung abscesses were found. The histopathological diagnosis was chronic focal suppurative pneumonia/bronchiectasis. Cholestasis was identified in the liver but was thought to be a late event, as the two biochemistry panels did not feature increased bilirubin. Gram stain and culture of lung tissue identified only a mixture of non-significant environmental flora, and a light growth of *Fusobacterium* sp. was cultured from a swab of the submandibular region. It was not clear whether the submandibular infection was related to the lung changes or the result of a separate event such as a penetrating wound. Exotic diseases were excluded and the investigation was closed.

A veterinarian called the exotic pest and disease hotline to report an unusual outbreak of haemolytic disease and Heinz body anaemia in a group of eight pregnant Thoroughbred broodmares. The horses all originated from a single property but had quickly deteriorated after being sent to a number of other properties. The outbreak occurred in October. Clinical pathology showed all mares to be mildly anaemic, with varying amounts of Heinz bodies (1–40 percent), and two of them had haematuria. The presence of Heinz bodies is considered abnormal in horses. Owing to the sudden onset of disease at the same time in all mares, a toxic rather than infectious agent was suspected. No toxins were found and no red maple was present on the original property. Several exotic agents can cause anaemia in horses, including equine viral arteritis (EVA), equine infectious anaemia (EIA) and leptospirosis. Disease in horses associated with *Leptospira* is uncommon, but this agent is well known for causing haemolysis in other animals. *Leptospira* serovars (including *L. Canicola*, which is exotic to New Zealand) were investigated by MAT. EVA titres were

slightly elevated in two mares (VNT titre 1:2; reference range 1:4+) and all EIA titres were negative. The foal of one affected mare was born dead, and postmortem confirmed fetal distress (meconium, squamous cell aspiration), but was otherwise unremarkable. Several other foals were born weak and the rest were apparently healthy. All of the mares fully recovered. Eight weeks after the initial tests, two of them were re-bled and again had low titres to EVA and various *Leptospira* serovars. There was no consistent pattern of elevation for any particular *Leptospira* serovar and no evidence of seroconversion. Based on the apparent point-source onset and other epidemiological factors, the most likely cause of the outbreak was undetermined toxicity. The multiple low-positive titres to *Leptospira* serovars and EVA were considered to be non-specific immunoreactivity, and this was supported by a lack of increase on paired titres. Heinz body anaemia in horses is rare and in this case no cause was identified. Exotic disease was ruled out by a combination of epidemiology and laboratory testing, and the investigation was closed.

A veterinary pathologist called the exotic pest and disease hotline to report oedematous limbs (dependent oedema) and mild diarrhoea in a 2-year-old colt in the Rangitikei district. Results of routine serology and haematology were unremarkable and no associated animals were reported to be ill. Serum samples tested negative for equine infectious anaemia virus by AGID, and for equine viral arteritis virus by VNT. The cause of the illness was not determined, but exotic causes were ruled out.

Equine viral arteritis ruled out

MPI investigators were notified by the AHL of a low positive titre (1:6) in the VNT for equine viral arteritis (EVA) virus in a horse undergoing routine pre-export testing. The horse was a healthy 4-year-old Thoroughbred gelding that had been imported from Australia in 2015 and was due to be exported to Hong Kong. Repeat testing of a sample collected 10 days later returned a negative result (1:2). The in-contact cohort of 17 horses in pre-export quarantine all returned negative EVA VNT results. Exotic disease was excluded and the investigation was closed.

MPI investigators were notified by the AHL that a 2-year-old Thoroughbred colt had tested positive to equine viral arteritis (EVA) virus in the VNT. However, the titre was very low (1:6) and suggested a non-specificity issue rather than actual exposure to the virus. The blood was re-tested and on this occasion determined to be negative (VNT titre 1:2 after < 4 days' incubation). New Zealand's horse population is free of EVA.

Equine piroplasmosis excluded

A veterinary pathologist notified MPI via the exotic pest and disease hotline of a 1-year-old horse with polyarthritis. Joint fluid aspirates and blood samples were tested by PCR for *Anaplasma phagocytophilum* and piroplasms including *Babesia* and *Theileria* spp. at the AHL (Wallaceville), with negative results. PCRs for *Borrelia burgdorferi* (the causative agent of Lyme disease) and *Rickettsia* spp. were also negative after being subcontracted to Acarus Laboratory, UK, for *Borrelia* PCR and University of North Carolina, VBDDL, USA for *Rickettsia* PCR. As exotic disease was excluded, the investigation was closed.

Brucella canis excluded

A veterinary pathologist notified MPI via the exotic pest and disease hotline of a 4-year-old dog with unilateral orchitis and epididymitis. The dog had just been castrated, and it had never left the South Island. Serum was submitted to the AHL (Wallaceville) for a *Brucella canis* card test, with negative results. The dog was in good health after being neutered, and with exotic disease excluded, the investigation was closed.

An 8-year-old dog that had been imported 4 years earlier from the UK had a history of chronic orchitis and epididymitis that was responsive to antibiotics. It was reported to MPI via the exotic pest and disease hotline by a veterinary pathologist. The dog had been castrated after a recurrence of inflammation, and pyogranulomatous epididymitis with testicular atrophy was noted on histopathology. A blood sample was tested at the AHL (Wallaceville) by a *Brucella canis* card test, with negative results. As exotic disease was excluded, the investigation was closed.

A veterinarian called the exotic pest and disease hotline to report a dog with a unilateral testicular mass. The 13-year-old dog had been imported from Australia a week prior, and had been booked in for castration. Although *Brucella canis* is not known to be present in Australia, testing was done to rule it out. The testicle was examined histologically and a *B. canis* card test and *Brucella* spp. PCR were conducted at the AHL (Wallaceville), with negative results. Multiple sex cord-stromal tumours (probable interstitial cell tumours) were identified by histology, with secondary partial ischaemic necrosis associated with thrombosis of blood vessels. There was no histological evidence of bacterial infection. Tumours of this type rarely metastasise and removal by castration is usually curative.

Canine babesiosis excluded

A veterinarian called the exotic pest and disease hotline to report a 9-year-old dog that had died while being treated for possible leptospirosis. Following the dog's death, test results for *Leptospira* spp. had been negative. The dog had been imported from Australia 5 years previously and had been very unwell and vomiting for a week, had jaundice, elevated liver enzymes and mild anaemia. These signs were consistent with leptospirosis, but also with blood-borne-pathogen infections such as babesiosis, ehrlichiosis and anaplasmosis. While these diseases are present in Australia and can have a chronic form, it was unlikely that they would be latent for 5 years before causing illness and death. The veterinary clinic had done a MAT serological test for *Leptospira* Copenhageni and a PCR of urine, and both were negative. However, there are several serovars of *Leptospira* present in NZ, and shedding of leptospiral organisms in the urine is intermittent, so these tests have a low sensitivity. Testing was done by the AHL (Wallaceville) to rule out exotic causes of death. Tests for nine *Leptospira* serovars, including the exotic *L. Canicola*, were subcontracted to the USA, and all were negative except a low positive for *L. ballum*, which is endemic in New Zealand. However, a much higher titre would be expected for an animal that had died of an infection with this organism, so it was considered that this low positive was caused by previous, routine vaccinations. Further

exotic disease tests also returned negative results, including IFATs for five *Babesia* spp., *Ehrlichia canis* and *Rickettsia* spp.; PCR for *Babesia* spp., and snap ELISA tests for Lyme disease (*Borrelia* spp.), *Ehrlichia* spp., *Anaplasma* spp., and heartworm (*Dirofilaria immitis*). The cause of the dog's death was not established, but as the most likely exotic agents were ruled out, the investigation was stood down.

Canine *Ehrlichia* excluded

A veterinary pathologist notified MPI via the exotic pest and disease hotline of a 10-year-old dog with a disease presentation consistent with exotic blood parasite infection. The dog had no history of travel. Some of the exotic diseases of concern are known to be spread by brown dog ticks, *Rhipicephalus sanguineus*, but the dog had had no contact with the area involved in a previous incursion of these ticks into New Zealand (since eradicated). Clinical signs were a generalised lymphadenopathy, fever and polyarthritis. Haematology and biochemistry results, including a lymphocytosis and hyperglobulinaemia, were consistent with a reactive inflammatory process. Severely decreased platelets were consistent with the tick-borne diseases, but at levels usually seen with immune-mediated thrombocytopenia. Cytology on fine-needle aspirates from multiple enlarged lymph nodes revealed reactive hyperplasia consistent with immune-mediated disease or blood parasite diseases. Cytology on joint fluid aspirated from multiple sites confirmed that immune-mediated disease was most likely, but an infectious cause could not be ruled out. An antinuclear antibody test was negative, meaning that systemic lupus erythematosus was unlikely, but this did not exclude other immune-mediated diseases. Joint fluid and blood samples tested negative at the AHL (Wallaceville) for *Ehrlichia canis*, piroplasm (including all canine *Babesia* spp.), and *Anaplasma phagocytophilum*. Further overseas subcontracted testing for rickettsial disease (including Rocky Mountain spotted fever, Lyme disease and *Bartonella* spp.) was postponed until the dog's response to treatment for immune-mediated disease could be evaluated. The dog responded well to treatment with corticosteroids and the thrombocytopenia and polyarthritis

resolved. As immune-mediated disease appeared to be the cause, the investigation was stood down.

Canine heartworm excluded

A Wellington-based veterinarian phoned the exotic pest and disease hotline to report coughing in a dog imported from Turkey and released from quarantine two weeks previously. The 2-year-old spayed crossbreed had been rescued with a broken leg, but had no other known health problems. The dog's coughing was described as soft, and more prominent after she had been lying down. Thoracic radiographs indicated a bronchiolar pattern most consistent with bacterial bronchitis (kennel cough). Because of the recent history of importation, *Dirofilaria immitis* (heartworm) and *Leishmania* spp. were tested for and ruled out at the AHL (Wallaceville). *D. immitis* ELISA and microfilaria concentration test (Knotts test) were negative, and an IFAT for *Leishmania* was also negative. The investigation was closed.

High-pathogenicity avian influenza and Newcastle disease ruled out

A member of the public in Canterbury contacted MPI via the exotic pest and disease hotline to report sudden deaths in his backyard chicken flock of 50 six-month-old chickens and 10 adults. The previous day all chickens had been well at morning feeding, but by evening nine of the younger birds were dead. The rest showed no indications of illness or disease. Highly pathogenic avian influenza and Newcastle disease were the main exotic differentials considered. Necropsies conducted at a regional veterinary diagnostic laboratory did not reveal any typical gross or histopathological lesions and excluded the involvement of an infectious process.

Duck mortality investigated

A fisherman notified the MPI exotic pest and disease hotline of 85 dead paradise ducks on a riverbank in the Manawatu district. No live ducks were seen, but other species of birds, including gulls, appeared to be healthy. It was arranged for the notifier to collect some of the freshest carcasses for post-mortem examination at Massey University. However, the carcasses were too decomposed to allow a gross or histopathological diagnosis. The

Incursion Investigator examined satellite photographs of the location of the dead birds, and saw pools and other areas of probable slow-flowing water that could have harboured *Clostridium botulinum*, the cause of botulism. It was considered that this was the most likely cause of the mortality event, and with no evidence of exotic disease and the event seemingly over, the investigation was stood down.

Rabbit syphilis excluded

A veterinarian contacted MPI to report an ulcerated soft tissue mass in the perineal region of a female rabbit. Rabbit syphilis (caused by *Treponema paraluischuniculi*) was suspected, though a foreign body or other bacterial infection was considered more likely. Bloods were taken and an IFAT test for *T. paraluischuniculi* was negative. This agent is not reportable and is widely present throughout the world's domestic rabbit populations, but has not yet been reported in New Zealand.

Exotic honey bee diseases excluded

A member of the public called the MPI exotic pest and disease hotline to report unusual behaviour in two bees that had flown into her Waikato house at night, circled the light, and were found dead the next morning. She had read about an emerging bee parasite, *Apocephalus borealis*, which has been described as causing disorientation or "zombie-like" behaviour in bees. *A. borealis* is a parasitic fly that has been found in North America but is not thought to be present in New Zealand. The bees were examined at MPI's Plant Health and Environment Laboratory, and determined to be honey bees (*Apis mellifera*) with no external or internal parasites. When bees are disturbed after dark they reportedly orientate to the nearest light source, and this may account for the unusual behaviour in this case. Exotic disease was ruled out and the investigation was closed.

European foulbrood excluded

A beekeeper in Napier had a disease in one of his hives and asked anASUREQuality Apicultural Officer to assist in the diagnosis. The disease was considered most likely to be halfmoon syndrome or residual parasitic mite syndrome, but these have the same signs

as European foulbrood (EFB), an exotic disease caused by *Melissococcus pluton*, so the Apicultural Officer notified MPI via the exotic pest and disease hotline. PCR testing for EFB in two larval samples was negative and the investigation was closed.

Exotic honey bee parasites excluded

A commercial apiary manager called the MPI exotic pest and disease hotline to report finding unusual mites in a hive from a 50-hive apiary. The mites were clustered together and attached to the backs of individual bees. Samples were collected and examined at the PHEL (Tamaki). A number of mite species were identified, including *Varroa destructor*, *Neocyphophthalmus novaehollandiae* and *Acarapis externus*. *V. destructor* and *A. externus* are both bee parasites, while *N. novaehollandiae* is associated with flowers and is frequently collected with pollen by bees. No notifiable species were identified and the investigation was stood down.

Exotic ticks excluded

A person in Carterton called the exotic pest and disease hotline after finding two ticks on his dog that had just returned from Gisborne. He was concerned that they might be brown dog ticks, *Rhipicephalus sanguineus*. An entomologist at PHEL (Tamaki) identified the ticks as *Haemaphysalis longicornis*, the endemic NZ cattle tick.

A tick was taken to a veterinary clinic in Wellington, from where it was reported to MPI via the exotic pest and disease hotline. It had been found on the floor of a house not long after a dog had visited from Otaki. Wellington is outside the usual home range of the NZ cattle tick, but the dog had recently been in rural areas, including north of Upper Hutt and on the Kapiti Coast. There was concern that the tick might be a brown dog tick, *Rhipicephalus sanguineus*, but it was confirmed as an engorged female NZ cattle tick, *Haemaphysalis longicornis*, by MPI's contracted entomologist. The investigation was closed.

A veterinarian called the exotic pest and disease hotline to report that an engorged tick had been found on a dog that was systemically unwell. The dog had a fever, white blood cell response and was generally poorly, though it had no history of overseas travel or contact with recently

imported dogs. A photograph of the tick was sent to a contract parasitologist, who identified it as *Haemaphysalis longicornis*, the NZ cattle tick. A request was made for the tick itself to be sent in for confirmatory identification, and for further details and samples from the dog, but these were not received. The investigation was stood down.

A member of the public from Motueka called the exotic pest and disease hotline to report finding two ticks on the family dog. The caller, who had moved to the area from Christchurch 10 days previously, was concerned that the ticks could be harbouring zoonotic disease. The tick was most likely to be the endemic New Zealand cattle tick (*Haemaphysalis longicornis*), which has been established in the Motueka area for more than 40 years. This species does not harbour any zoonotic diseases. However, an exotic differential was the brown dog tick (*Rhipicephalus sanguineus*), of which there was an incursion in Canterbury in 2015 (Pleydell *et al.*, 2015). The notifier was able to provide an MPI Incursion Investigator with digital pictures of the ticks, from which an entomologist confirmed the ticks were *H. longicornis*. The investigation was closed.

A Christchurch veterinary clinic called the exotic pest and disease hotline to report finding 13 ticks on two dogs. The endemic New Zealand cattle tick (*Haemaphysalis longicornis*) is not known to be widely established in the Canterbury region, nor does it usually target dogs. An exotic differential for ticks on dogs is the brown dog tick (*Rhipicephalus sanguineus*), of which there was an incursion in Canterbury in 2015 (Pleydell *et al.*, 2015). At the request of an MPI Incursion Investigator the ticks were submitted to the PHEL (Christchurch), where they were confirmed as being the New Zealand cattle tick. It is most likely that the dogs had picked up the ticks when recently visiting rural properties in Northland, where the NZ cattle tick is widely established. An exotic tick incursion was ruled out and the investigation was closed.

A member of the public from Nelson found ticks on his dog and called the exotic pest and disease hotline. The notifier thought these ticks might be the exotic brown dog tick, *Rhipicephalus sanguineus*. Photographs of the ticks were

forwarded to an MPI-contracted tick expert, who confirmed them as the NZ cattle tick, *Haemaphysalis longicornis*. This species has been established in the Nelson region for more than 40 years. An exotic tick incursion was ruled out and the investigation closed.

A member of the public from the Tasman region called the exotic pest and disease hotline to report having found a tick where her dog slept. Following a Google search she had become concerned that it could be the exotic brown dog tick (*Rhipicephalus sanguineus*). She had lived in the region for 17 years with the dog and this was the first time she had encountered a tick. The dog was not imported and had no known contact with imported dogs. The tick was submitted to the PHEL (Christchurch), where it was confirmed to be a New Zealand cattle tick (*Haemaphysalis longicornis*). An exotic tick incursion was ruled out and the investigation was closed.

A member of the public called the exotic pest and disease hotline to report finding a tick on the floor of his house. He was concerned that it might be a brown dog tick (*Rhipicephalus sanguineus*). There had recently been several overseas guests in the house and the notifier had never previously found ticks in his home. The tick, an adult female, was identified as the endemic New Zealand cattle tick (*Haemaphysalis longicornis*), found widely within New Zealand. An exotic tick incursion was ruled out and the investigation was closed.

A dog owner in Auckland found an engorged tick on her kitchen floor and called the exotic pest and disease hotline. The dog had been staying at Muriwai on a lifestyle property with farm animals a couple of weeks previously, and had most likely picked up the tick there. Photographs of the tick were sent to MPI's contracted entomologist, who tentatively identified it as a NZ cattle tick, *Haemaphysalis longicornis*. The identification was confirmed after the tick was sent in for examination, and the investigation was closed.

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Quarterly report of investigations of suspected exotic marine and freshwater pests and diseases

Toheroa disease investigated

A researcher at Waikato University called the MPI pest and disease hotline to report the presence of blisters between the periostracum and the shell on a large number of toheroa (*Paphies ventricosa*) on Ninety Mile Beach (Figure 1).

The researcher suspected that the blisters might be associated with past mass mortalities.

Toheroa are large endemic bivalves found in the surf zone of sandy beaches on the west coast of Northland, the west coast north of Wellington, and the south coast of Southland. They were once abundant, but were intensively harvested during the first half of the twentieth century, and most populations declined to low levels. All commercial harvesting ceased in 1969 and the recreational take has been tightly regulated ever since, to help populations recover.

MPI acquired a special permit to collect toheroa to investigate whether a disease was the cause of the blistering. Ten animals were submitted to the MPI Animal Health Laboratory (AHL) at Wallaceville for diagnostic testing (histological, bacterial, and PCR to identify species).

Bacterial isolation found several species of environmental bacteria that are common in marine environments, including *Rahnella*, *Acrobacter*, *Pseudomonas* and *Vibrio* spp. These are usually opportunistic pathogens that readily invade animals that are already immunocompromised.

A *Rickettsia*-like organism (RLO), another very small bacterium, was also present in every specimen submitted. RLOs are a complex taxonomic group, difficult to identify to species level. Like other widespread environmental bacteria they can cause disease in marine organisms.

However, in this case the MPI fish pathologist determined that the blisters were consistent with gas-bubble disease (Malouf *et al.*, 1972). When water becomes supersaturated with nitrogen or oxygen, the excess gas can force its way into the shells and tissues of marine organisms. In this case, it is thought

Exotic marine pest and aquatic disease investigations are managed and reported by MPI Diagnostic and Surveillance Services, Wallaceville. The following is a summary of investigations of suspected exotic marine diseases and pests during the period from January to March 2017.

that a sudden temperature rise when the cold seawater hit the sun-warmed tidal shore of Ninety Mile Beach in Northland, may have caused oxygen supersaturation. Disease was ruled out and the investigation was closed.



Figure 1: Toheroa (*Paphies ventricosa*), showing blisters between the periostracum and the shell

Strawberry-like mass on Wellington beach

A member of the public called the MPI pest and disease hotline to report a mass of strawberry-like clumps washed up on a Wellington beach. An Incursion Investigator arranged for a sample to be collected and submitted to the Marine Invasive Taxonomic Service (MITS) at NIWA for identification. An ascidian taxonomist identified it as *Aplidium adamsi*, a colonial sea-squirt native to New Zealand. As there was no biosecurity risk, the investigation was closed.

Mussel deaths investigated

A reporter from the *Westport News* contacted the Department of Conservation (DoC) about a large number of dead mussels at Cape Foulwind, southwest of Westport. DoC contacted the AHL via the exotic pest and disease hotline, and samples were collected and sent to the laboratory. The mussels tested negative for the parasite

Perkinsus olseni. Cysts of the polychaete *Nematopsis* sp. were found, but these are often seen in mussels and were not considered to be of pathological significance. Three out of 10 mussels were found to have *Rickettsia*-like organisms in the gills or the digestive gland, and three out of 10 also had apicomplexan “X” (APX) cells present. Four mussels showed an immune response in the form of focal haemocytes, brown cells and melanin, and some rod-shaped bacteria were present. This low prevalence of several different potential pathogens indicated a lack of any single causal pathogen, so these mussels were considered to have died from multiple causes. No biosecurity risk was identified, so the investigation was closed.

Koura mortality event

In January 2017, MPI was alerted to a news article referring to a mass mortality of koura or freshwater crayfish (*Paranephrops planifrons*) in a Waikato stream during December 2016. No other species were reported to have died. The notifier, a veterinary epidemiologist, suggested the cause of the mortality could be crayfish plague (*Aphanomyces astaci*), an exotic pathogen that has largely destroyed populations of European crayfish across Europe. *A. astaci* is an aquatic mould that only parasitises freshwater crayfish, and is notifiable to the World Organization for Animal Health (OIE). New Zealand is considered free of the disease. The Waikato Regional Council (WRC) investigated the event, collecting water samples and dead koura specimens to see if there was any environmental or toxicological cause. MPI Incursion Investigators visited the stream but did not notice any further mortality of koura or other aquatic life. Together with the WRC freshwater ecologist, they agreed

that the stream had healthy koura, eel and trout populations of all age groups. The koura specimens were sent to the AHL for pathogen testing but all tested negative for *A. astaci* by molecular diagnostics, so crayfish plague was ruled out. There was no indication of any infectious cause, and no further reports of sick or dead koura were received, so the investigation was closed.

Algal mats washing ashore in Auckland

A member of the public called the MPI pest and disease hotline to report large, thick mats of what appeared to be algae washing ashore at the south end of Long Bay, Auckland. The caller was concerned as he had never seen such algal mats in the 40 years he had been living in the area. Photos of the beach were submitted and chilled samples of the algae were sent to MITS, but unfortunately they were too decomposed for identification. There appeared to be clumps of the red algae *Jania* and *Hypnea* and, from the photos, there also appeared to be a brown seaweed present, *Cladostephus* sp., which can form an entangled mass that dislodges and collects other marine species as it washes ashore. These masses are similar in appearance to those described by the caller. It is unknown what caused the algal bloom in Long Bay, though algae blooms typically occur when growing conditions become favourable, e.g. warmer weather, and increased sunlight and nutrient availability. The bloom can then die off suddenly when those conditions change, which may result in algal mats being washed ashore and causing a nuisance. No exotic or non-indigenous species appeared to be involved, so the investigation was stood down.

Unusual white balls in snapper

A recreational fisher called the MPI pest and disease hotline to report unusual growths in snapper (*Chrysophrys auratus*). He reported strange white fatty balls in the mid-section of the fish. The fisher reported that one out of four fish was affected, and that he knew of other people who had seen this in fish caught from the same area (Army Bay, Whangaparaoa). There are two conditions that present similarly in snapper from this area, and both have been reported for a

number of years. One is hyperostotic pterygiophores, caused by a type of fungus. The other has not yet had a causal agent isolated and is described as “cysts of unknown aetiology” (Hine & Anderson, 1981; Roubal, 1994; McClean *et al.*, 1987). After examining a snapper carcass sent to the AHL, a fish pathologist was satisfied that this was a case of the latter condition. This is not thought to pose a biosecurity risk and the investigation was closed.

Catfish investigation

Contractors working on an Auckland stream realignment found an unusual fish, which they photographed and released. A freshwater ecologist overseeing the work was alerted to the discovery and notified MPI via the pest and disease hotline. An Incursion Investigator who examined the photos was able to confirm that it was clearly a catfish species, but the photos were inadequate to identify the genus. The Import Health Standard for Ornamental Fish (FISORNIC ALL) permits the importation of many species of catfish into New Zealand, provided they undergo quarantine before clearance. From the markings on this specimen, it may have been a janitor fish (*Pterygoplichthys* sp.) or a suckermouth catfish (*Hypostomus* sp.). This individual was likely to have been released from an aquarium, rather than being from a wild breeding population. The freshwater ecologist monitors the stream every few months and will notify MPI if more catfish are found. As no biosecurity risk could be confirmed, the investigation was closed.

Eel mortality investigation

MPI was advised by the Gisborne District Council that a landowner had reported more than 20 dead eels in a tidal section of the Taruheru River. A single eel was collected and sent to the AHL. No abnormalities were seen when it was examined, and molecular testing was negative for eel virus European X (EVEX) and picornavirus, both of which have been found in eels in New Zealand (van Ginnekan *et al.*, 2004). The Council reported that this mortality event had been preceded by a dry spell that was followed by a weekend of heavy rain (20–40 mm), so it is likely that these environmental factors contributed to the mortality, possibly by disturbing stagnant water that in turn flowed down the

waterway, affecting the eels. No further mortalities were observed. The Council was informed and the investigation was stood down.

Dead bullies in Lake Taupo

The Waikato Regional Council (WRC) notified MPI of a report of numerous dead cockabullies in the Waitahanui River. WRC sent out freshwater scientists and environmental monitoring staff to investigate the report. A hundred or so common bullies or gobies (*Gobiomorphus cotidianus*) were found dead where the stream flows into Lake Taupo. No other freshwater species were affected. WRC environmental monitoring staff found that the water temperature was slightly elevated and there was increased sedimentation and turbidity from recent heavy rain. Freshwater ecologists considered that these potential environmental stressors, coinciding with the common bully's spawning season, were the most likely cause of the mortality event. As there was no evidence to suggest that disease played a role in this incident, the investigation was closed.

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Exotic plant and environment investigations report: January to March 2017

New to New Zealand organisms: positive reports

The new to New Zealand truffle species *Tuber dryophilum* was reported by a Plant and Food Research (PFR) scientist after it was found at three Canterbury locations: PFR's experimental plots at Lincoln and two separate West Melton sites. Truffles are the fruiting bodies of mycorrhizal fungi that live in a symbiotic relationship with numerous trees. In New Zealand these are typically oak, pine, poplar and hazelnut. No New Zealand native tree species have been found to host truffle-forming *Tuber* fungi. Commercial production consists only of harvesting truffles from beneath orchards (truffières) of host trees. To date, all finds of *T. dryophilum* have been among commercial plantings of *T. borchii* (bianchetto truffle), where they are considered a contaminant, reducing yields of *T. borchii*. As *T. dryophilum* is common where *T. borchii* grows in Europe, it is possible that it entered New Zealand when *T. borchii* inoculum was first imported in about 1985, or during later importations that occurred until at least 2006, up to which time taxonomic discrimination between truffle species was largely based on morphology. A similar contaminant, *T. bromale*, was the subject of a biosecurity response in 2006 and was concluded to have likely entered through this pathway. Confusingly, *T. dryophilum* was previously considered present in New Zealand, based on specimens held by Landcare Research (LCR). However, a recent molecular and morphological review of *Tuber* spp. held in LCR and PFR collections (Bulman *et al.*, 2010), reassigned these specimens to *T. maculatum*, a species already considered present in NZ. Truffle material submitted to MPI has been validated as *T. dryophilum*, and specimens have been lodged in the New Zealand Fungarium collection. An evaluation was made to assess the threat posed by this species and sent for comment to PFR and scientists who support the New Zealand truffle industry. The scientists supported MPI's view that,

The Ministry for Primary Industries' (MPI) Incursion Investigation teams and the Plant Health Environment Laboratory (PHEL) teams investigate and diagnose suspect exotic pests and diseases in the plant and environment sectors. Investigators and scientists are based in Auckland and Christchurch. These teams provide field investigation, diagnostic testing and technical expertise on new pests and diseases affecting plants and the environment. They also have surveillance and response functions and carry out research and development to support surveillance and incursion response activities.

although this species does impact our truffle industry, it is already widespread here and we cannot easily detect and eradicate it, so an eradication attempt would have a low likelihood of success and low cost/benefit.

A Plant and Food Research scientist advised that the truffle *Tuber menseri* had been identified by an independent diagnostic mycologist, based on a single fructification collected on an Invercargill property. The specimen was received along with an additional species, *T. dryophilum*. While *T. dryophilum* is now considered to be established in NZ, there is uncertainty as to the status of *T. menseri*. To date the species remains undescribed and the taxonomic status is unclear. While the DNA evidence provided to MPI has been confirmed as originating from *T. menseri*, the original specimen has been lost and there are no voucher specimens for comparison. Furthermore, the truffière from which it was obtained no longer exists: all the host plants have been removed and destroyed, and the land has been converted back into pasture. Hence the plant host species of the truffle cannot be validated. Bonito *et al.* (2010) reported an association between *T. menseri* and *Pinus sylvestris* and *Picea abies* in New Zealand (see also Bulman *et al.*, 2010), but the origin of the sample material used in that study could not be validated from any New Zealand source. The sum of substantiated evidence is considered insufficient to consider *T. menseri* as definitely established in New Zealand. Collated evidence will remain in MPI's database

pending further data from new truffle collections or other sources.

MPI received a report of a moth, suspected to be the exotic guava moth *Ophiusa disjungens*, from NatureWatch (<http://naturewatch.org.nz/>), an online tool for citizen science and community-based monitoring projects on nature and the environment. The identification was subsequently confirmed by MPI. The observer confirmed that the moth was caught at Mission Bay, Auckland on 15 December 2016. Guava moths are native to Southeast Asia, but are also present in Australia, Hawai'i and several Pacific Islands. *O. disjungens* feeds on eucalyptus and guava and has been reported feeding on *Metrosideros polymorpha* though it is not known whether it can complete its life cycle by feeding on this plant. It appears to pose a low biosecurity risk in Hawai'i. High Risk Site Surveillance Programme staff have been told to look out for the caterpillars, but have not yet found any. *O. disjungens* is described in the literature as a "fruit piercer" but its harm is largely limited to guava and it causes more damage by defoliation. As the specimen found was a male, and a population has not been found, its establishment in New Zealand appears unlikely.

Biosecurity pests and contaminants

While unpacking a kitset of wooden outdoor furniture made in Vietnam and purchased from Briscoes in Palmerston North, a live insect was found in the packaging and a mud daub

was noticed attached to the wood. The insect was identified as *Inopus rubriceps*, the Australian soldier fly, which is established in New Zealand and was likely to be a local contamination. The wasp nest was empty and therefore of no biosecurity risk.

Pumpkin kernels purchased about three months previously were found to be bunched together in clusters by a web-like, sticky material. Several clusters looked as though the whitish substance was actually coming out of the kernels. The notifier said it did not look as though roots were emerging from the seeds. From a photo it was determined that they were infested with *Plodia interpunctella*, the Indian meal moth. Pupae were evident and there was larval frass among the kernels. *P. interpunctella* is a common storage pest and is established in New Zealand.

A live spider was found in a store that retailed imported goods. The person reporting said the spider was large and unusual in appearance, and possibly from overseas. Photos of the spider were shown to an MPI entomologist, who identified it as the large brown vagrant spider *Uliodon albopunctatus*. This is an endemic species common in both the North and South Islands, and not a biosecurity risk.

The pharaoh ant *Monomorium pharaonis* was found at Mt Maunganui seaport through the National Invasive Ant Surveillance Programme. This species has been established in New Zealand since the 1950s and has become an urban pest in hospitals and commercial premises. No action was taken.

A notifier imported several boxes of straw hats from China. The boxes were dry on the outside, but the hats wrapped in plastic bags were completely soaked. White fungal mycelium and orange structures (suspected to be spore bodies) became visible on the hats after a week. Samples were submitted to MPI's Plant Health and Environment Laboratory and confirmed as *Neurospora* sp. Fungi in this genus are considered environmental contaminants and pose no biosecurity risk.

A caller to the exotic pest and disease hotline reported finding a spider in a packet of green grapes imported from California and bought from a Blenheim supermarket. The caller believed it

was a black-footed yellow sac spider (*Cheiracanthium inclusum*), an exotic species, and said it had an egg sac. This identification was subsequently confirmed by MPI's Plant Health & Environment Laboratory but considered to be a solitary hitch-hiker. The spider was destroyed and no further action was taken.

Canterbury Museum staff advised MPI that a grey huntsman spider (*Holconia immanis*) had been found in imported clothing at a local sportsgoods shop. It had possibly hitchhiked on a recently imported consignment of 13 small boxes of clothing. All the cardboard boxes were thoroughly checked and no more spiders were found. While this was a solitary detection on low-risk goods, the detection was turned into a media story (<http://www.stuff.co.nz/national/89514340/Bio-security-systems-under-threat-as-trade-increases?cid=app-android>). This demonstrated a lack of understanding of the biosecurity system, which is a risk-based system addressing biosecurity risk within multiple layers (including border and post border layers). Inevitably, low-risk organisms on low-risk goods do slip through the border occasionally and the Post Border team is the layer that deals with such cases, as it successfully did on this occasion.

A gardener found large numbers of small invertebrates that appeared to be attacking the tubers of home-grown potatoes. Examination of photos was inconclusive and a sample was requested for formal identification. The invertebrates identified as spotted snake millipedes, *Blaniulus guttulatus*, a species established in New Zealand.

A person reported finding little black insects emerging from a bunch of Chinese garlic three days after purchase. Another bunch of Chinese garlic of a different brand that he also had at home was free of insects. A sample was requested and the insects were identified as onion aphids, *Neotoxoptera formosana*, a species already present in New Zealand.

A member of the public handed an unusual wasp to MPI officers in Whangarei. From photos it was suspected to be a yellow paper wasp, *Polistes olivaceus*, an unwanted organism not known to be present in New Zealand, and this was confirmed by PHEL (Tamaki). The wasp was also noted to be a queen with some ovary development but not

enough to suggest it had established a nest. The wasp was caught at a hardstand area much used by visiting foreign vessels for repairs and maintenance. A quarantine inspector was instructed to inspect the area and found two paper wasp nests, but these were identified as *P. chinensis*, a species established in New Zealand. An Incursion Investigator and MPI entomologist carried out further inspections but did not find any wasps. Enhanced passive surveillance of the area was also carried out. Stakeholders including the Northland Regional Council and the Department of Conservation were notified. No public notifications of suspect yellow paper wasps were received from the passive surveillance campaign. The Port Whangarei Marine Centre was asked to be vigilant for this wasp and to notify MPI of any further finds. The site was also targeted for extra surveillance through the High Risk Site Surveillance Programme. The case was closed as this was considered to be an isolated hitchhiker and there were no further finds.

Lake snow, *Lindavia intermedia*, was reported by Otago Regional Council on 19 January 2017. Lake snow is the common name for a diatom present in several Queenstown lakes and causing nuisance by creating gelatinous masses that foul fishing equipment and pipes. Research shows the organism has likely been present in New Zealand since at least 2004, but has been increasing in density more recently. The cause of the proliferation is unclear but may be related to increased nutrient inputs, climate change and the recent establishment of the water-flea *Daphnia pulex*. A response was initiated and a report was prepared recommending no immediate action to eradicate or contain lake snow.

There have been numerous reports of brown crazy ant *Paratrechina longicornis* at Ports of Auckland during the reporting period, largely through the National Invasive Ant Surveillance (NIAS) Programme. The first report was on 20 January 2017. A work instruction was issued to Flybusters Ltd to eradicate the ants, and they visited the site on 25 January. The ants had been found close to a concrete barrier adjacent to a weighbridge. Toxic bait was laid and a medium-sized nest with about 300–400 workers was found. The area was sprayed with a non-repellent residual

insecticide and toxic bait stations were laid alongside a long-term pitfall trap. There was a follow-up visit on 31 January to determine whether this had eradicated the nest, and no ants were seen. A third visit on 9 February again found no ants and the case was closed.

More brown crazy ants (*Paratrechina longicornis*) were found at Ports of Auckland through the NIAS Programme. On 9 February the ants were found at a light pole on Jellicoe Wharf in a pitfall trap (also called a dome trap), a relatively new device being trialled this season in conjunction with the usual baited traps. The pitfall trap is designed to attract foraging ants over long periods (up to three weeks), and kills the ants when they enter, whereas the usual baited traps only work for two hours and do not kill ants that enter. A Work Instruction was issued to Flybusters Ltd to eradicate the ants. On 31 January a single specimen of *P. longicornis* was found in the trap, which was then left for a further week to see what other species might be caught. On 9 February more brown crazy ants were found in the trap, indicating a nest nearby. The trap was left in place as it was not scheduled to be removed until 21 February. Fresh toxic bait was laid but attracted no more ants. Prophylactic baiting in the area had occurred recently and there was one dead brown crazy ant at this bait. It was considered likely that the ant nest had been destroyed by the prophylactic baits. On 21 February no ants were seen in the trap or nearby. A final precautionary check was undertaken on 6 March and no further ants were seen. The area was further treated with a precautionary spray and more toxic bait was laid. The case was then closed.

Customs at the Ports of Auckland notified MPI border staff after finding four ants crawling on their X-ray machine. The ants did not resemble any local species and were subsequently identified as *Pheidole* sp. by PHEL (Tamaki). Movement control and a contact insecticide were deployed immediately. Flybusters Ltd carried out an additional spraying of the building and conducted a site investigation with monitoring traps. No further ants were found. Flybusters Ltd rechecked some of the NIAS traps in the vicinity from the previous week and found no exotic ants. These traps were used to survey the area outside the Customs building as part of the surveillance programme. Species

identification could not be done with molecular tools. The investigation was closed owing to absence of exotic ants in the traps and the precautionary spray treatment in the area. Further surveying will be carried out via NIAS.

There was yet another find of brown crazy ants (*Paratrechina longicornis*) at Ports of Auckland through the NIAS Programme on 15 February. A Work Instruction was issued to Flybusters Ltd to eradicate the ants, which were found adjacent to a concrete barrier. Flybusters visited the site on 6 March and laid toxic bait stations and delivered a precautionary spray treatment. No exotic ants were seen. On a further visit on 16 March to assess the efficacy of treatment there were no signs of ants, the toxic bait stations were replenished and the case was closed.

A fourth detection of brown crazy ant (*Paratrechina longicornis*) was made at Ports of Auckland through the NIAS Programme on 23 February. More than 20 worker ants were found in the baited trap. A Work Instruction was issued to Flybusters Ltd to eradicate the incursion, which was found at the base of a light pole. Flybusters Ltd visited the site on 6 March and put out toxic bait. A medium-sized nest containing more than 300 workers was found and a precautionary spray treatment was applied. The site was revisited on 16 March and no ants were seen. Toxic baits were replenished and the case was closed.

More brown crazy ants (*Paratrechina longicornis*) were found during the NIAS Programme in a long-term pitfall trap at Tinley Street, Ports of Auckland, on 16 March. The trap was in an area not covered by the three prophylactic baiting treatments laid by NIAS. On 23 March the nest was located after attractant baiting around the perimeter of a building, where dirt had accumulated and weeds had built up. The nest size based on worker activity was very small (likely a single queen with no more than 25–30 workers). Toxic baits and precautionary spray treatments were applied to the nest and base of the building and adjacent barriers. On 28 March fresh toxic bait was applied but no more ants were seen in the vicinity so it was concluded that the ants had been destroyed. No further action was recommended, but this area will be routinely re-surveyed during the NIAS 2018 season.

Ghost ants (*Tapinoma melanocephalum*) were found at the CSL Container Park Transitional Facility in Auckland through the NIAS Programme on 24 February. More than 30 workers were found in a baited trap near the edge of an open-sided workshop. A Work Instruction was issued to Flybusters Ltd to eradicate the ants and they visited the site on 15 March and laid out toxic bait. A small nest was found in a gap between two wooden blocks and a second nest was found under a sheet of plywood two metres away. Nest size (based on worker activity) was determined to be medium (likely with multiple queens and more than 300 workers). A third nest was also found in a drainpipe flashing. Precautionary spray treatments were applied and toxic bait stations laid. When the site was revisited on 22 March all the nests had been eradicated, but the toxic bait stations were replenished.

A nest of ants of the new to New Zealand genus *Tapinoma* was found at the Port of Timaru through the NIAS Programme on 14 February. The nest was found at the container terminal on the main wharf, in an storage area for refrigerated containers. It was in a 30-cm cavity full of dirt, rubbish and discarded electrical equipment. A Work Instruction was issued to Flybusters Ltd, who visited the location on 23 February and placed toxic bait within a 50 metre radius from the find. The nest was treated with insecticide and a precautionary spray treatment out to 20 metres was applied. Toxic bait stations were also laid. A follow-up visit the following day yielded no further ants and no further signs of nest activity were seen, so the case was closed.

An ant colony was found in Whitianga in a pile of packaging material, some of which had only recently arrived in New Zealand from Bali. The colony was considered to be possibly an exotic species. The volume of packaging material (cardboard and wooden crates) was too large to easily treat or dispose of. However, it was stored in a shipping container, so ant specimens were collected and the container was closed and sealed in order to contain the risk until samples submitted to PHEL (Tamaki) could be identified. The door seals, vents and surrounding ground were also treated with insecticide as an added precautionary measure. The ants were subsequently identified as the black house ant *Ochetellus glabror*, a species

already established in New Zealand and common in Whitianga, so it is likely they were of local origin.

Ants were noticed at various locations around a Transitional Facility in Paremata, near Wellington. They were an unusual colour and their appearance coincided with the arrival of a devanned consignment from the US. Samples were couriered to PHEL (Christchurch), where they were identified as *Monomorium antarcticum*, an endemic species widely distributed in New Zealand.

The packaging material with a book received from an online bookshop in the UK was found to contain a lot of ants. When the book was removed from the packaging and shaken onto the deck outside the house, more ants were seen. They were identified as the black house ant *Ochetellus glaber*, which is established in New Zealand but often found in imported goods as it is widely distributed throughout the world. It commonly nests within the structure of buildings and could have been picked up in the post within New Zealand.

A member of the public found many ants and eggs in his telephone when changing the batteries. The telephone, from Vietnam, had been bought more than six months previously and was in operation. The notifier had seen ants in the vicinity during the previous couple of months. They were identified by PHEL (Tamaki) as the little black mono ant *Monomorium fieldi*. This organism is present in New Zealand and so is not a biosecurity issue.

Staff at a car parts dealer found numerous ants and eggs in a car body imported from Australia. They treated it with fly spray and contacted MPI. The car had already been stored at the premises for six months, so the infestation was most likely to be a local species. Samples were requested to rule out an exotic infestation and were identified as the white-footed house ant *Technomyrmex jocosus*, a species that is established in New Zealand and is of no biosecurity concern.

Suspect forest and timber pests

A caller found boring insects in two wooden giraffe statues imported from South Africa about 9 months previously. Photos appeared to be consistent with bostrichid beetle, and a sample was

identified as *Heterobostrychus brunneus* (auger beetle), an exotic species present only in Africa. The two statues and another carving from the same retailer in South Africa were heat-treated to the MPI-approved standard.

An insect sweep-net purchased from an Auckland discount variety store was found to have a bamboo handle heavily infested with an insect borer, subsequently identified by MPI as *Chlorophorus annularis*, a longhorn beetle considered absent from New Zealand. MPI visited the store and found only a single net remaining for sale. No borer were found after it was bought and examined. However, identical nets were for sale in a different variety store in the same shopping mall. In all cases the bamboo handles were shrink-wrapped in a permanent plastic coating, obscuring visual evidence of any borer present. To estimate the scale of this interception MPI bought 10 nets and examined the handles. Seven nets contained live *Dinoderus minutus* borer and previous borer damage was evident in all the remaining three. The entire stock of nets at the store was purchased by MPI and destroyed to mitigate the biosecurity risk. Although *C. annularis* is known to attack bamboo that has a low moisture content (e.g., harvested and drying bamboo), there are no records of it causing economic damage to food and other economically important crop plants. *D. minutus* is commonly intercepted in goods imported into New Zealand – mostly bamboo, but sometimes softwood items. It is a much smaller beetle than *C. annularis*, with correspondingly smaller larval feeding galleries and beetle exit holes. Neither species is considered present in New Zealand. Both remain regulated by MPI despite arguably not posing any significant biological or economic risk.

A retail store owner reported live borer in wooden incense holders originating from India and purchased from a New Zealand importer/wholesaler. MPI visited the store and collected 103 incense holders. Six of these were destructively sampled and two were found to contain live *Dinoderus minutus* larvae in feeding galleries (tunnels). Some of the incense holders had borer exit-holes and internal tunnelling, but no live larvae. Dead larvae were also found, suggesting that they had been treated, but to a poor standard. The remaining incense holders

were assessed for borer exit-holes. Traceback to the importer revealed that they had been imported in late 2015. No stock remained with the importer, the product line had been discontinued and no further goods had been imported from the supplier in India. Although the particular consignment could not be traced, the MPI database showed that there were heat-treatment certificates for all declared wooden goods imported by the company. *D. minutus* is frequently intercepted at the border in wood and, more commonly, bamboo goods, and is considered a low biosecurity risk. The immediate risk was addressed by freezing the remaining stock. No further action was considered necessary.

A person contacted MPI via the pest and disease hotline after noticing sawdust falling from a wooden coat hanger, one of about 200 bought from a warehouse in Rotorua on three separate occasions. The coat hanger was destructively assessed by PHEL (Tamaki) and one dead adult and one live larva were found. Identification was confirmed as the Oriental wood borer or lesser auger beetle, *Heterobostrychus aequalis*, which is a pest of timber and not present in New Zealand. All the remaining coat hangers were sent to PHEL (Tamaki) and treated by freezing for 10 days. The infested coat hanger had been purchased several months previously, suggesting that all stock from the same consignment had likely already been distributed, so the investigation was closed.

Suspect exotic termites were reported by a Wellington-based pest-control operator. They had been seen among swarming adults of the New Zealand native termite *Kalatermes brouni*. The four adult termites seen were considered so different in size and colour to the alate *K. brouni* that they might be another species. However, specimens received by MPI were confirmed as *K. brouni*. The cause of the observed differences remained unclear.

Suspect agricultural and horticultural pests

Suspect fruit fly were found in a recently arrived refrigerated sea container in Invercargill. The container had entered through Bluff Southport from Australia on 3 March. It had been cleaned, inspected, stored for 10 days, then transported by truck to South Pacific

Meats' processing facility in Awarua, Invercargill, to be packed with frozen meat and returned to Southport for export. On arrival at Awarua the truck driver noticed a plastic bag in the externally accessed refrigeration unit floor of the empty container. The bag contained maggots and pupae associated with a rotten banana. MPI Verification staff reported the event to local MPI border staff. The risk material was placed into a sealed bag and couriered to PHEL (Christchurch). Next day the lab confirmed that the bag was negative for high-risk tephritid fruit fly. Subsequently the insects were confirmed to be *Drosophila melanogaster*, the common vinegar fly, a species established in New Zealand.

Two live suspect brown marmorated stink bug, *Halyomorpha halys* (BMSB) were picked up at an Auckland Transitional Facility dealing with air freight from China. These could not be associated with any goods that were recently imported. The MPI border team inspected the premises, collected samples and fumigated the five suspect containers containing 5 000 pallets of clothing (non-risk goods). The sample was identified as *Cermatulus nasalis*, a species of shield bug established in New Zealand that looks similar to BMSB.

Small holes were noticed on the skin of navel oranges imported from the US and purchased in Kerikeri. When the fruit were cut open, small live caterpillars were found to be feeding under the skin. Examination of samples provided to PHEL (Tamaki) determined the larvae were guava moth *Coscinoptycha improbana*, a common insect found attacking garden fruit plants in northern New Zealand. It appears likely that the imported oranges were contaminated with guava moth larvae while sharing a fruit bowl with home-grown fruit. Larvae and pupae of the common vinegar fly *Drosophila melanogaster* were also found on the oranges submitted to MPI. Molecular testing of these specimens ruled out *D. suzukii*, a high-priority pest absent from New Zealand and of significance to New Zealand horticulture.

A Japanese importer of marrowfat pea seed exported through an Ashburton seed-dressing company reported finding a suspect weevil in the consignment. Photos submitted by the importer were not clear enough to identify the insect to

species level but an interim identification of a bean weevil, either *Callosobruchus chinensis* or *C. maculatus*, was made by MPI. The Japanese importer was asked to send a clearer photo and engage an entomologist to identify the insect. The marrowfat pea crop had been grown in the Methven/Cairnbrae area from seed supplied by a local seed producer. The pea crop had been harvested in February 2016 and delivered directly to the seed-dressing company, dressed and bagged in 50-kg polypropylene sacks. In August 2016 a 21.5-tonne shipment was sent to Yokohama, where it was unloaded onto pallets in a Japanese warehouse before being transferred directly to cool storage. The grower had the pre-shipment seed certification samples in their Ashburton laboratory and when these were examined no signs of contamination were found.

The Japanese importer also imports peas from Canada and the UK. Representatives from both the company that provided the original seed and the grower visited the Japanese importer and inspected the warehousing and storage facility. They also obtained a copy of an identification report, which was submitted to PHEL (Tamaki). From the photos in the report, the PHEL entomologist considered the beetles were likely *Callosobruchus chinensis* (Oriental cowpea beetle), a species not known to be present in New Zealand. (This diagnosis was partly based on the assumption that all three beetles in the photos were females.) The Japanese report did not, however, identify the organism beyond the bean weevil family (Coleoptera: Bruchidae). *C. chinensis* is a pest of harvested peas and is established in Canada and Japan but absent from NZ, so this case was determined to be a local contamination within the Japanese warehouse and not associated with marrowfat pea seed imports from New Zealand.

On 22 February 2017 an Auckland shipping company employee spotted a suspect brown marmorated stink bug *Halyomorpha halys* (BMSB) outside the glass window on the 11th floor of the Rabobank building. He took photos of the bug but was unable to catch it as it was outside and inaccessible. MPI entomologists examined the photos, but owing to the poor quality and the fact that the insect was photographed on the ventral side, diagnosis was

inconclusive though BMSB could not be ruled out. The specimen showed some key morphological characters of BMSB, but in the absence of a picture from the other side it could also be the native brown soldier bug *Cermatulus nasalis*, or another exotic stink bug, *Rhaphigaster* sp. The notifier was asked to send some more pictures with a ruler on the side for scale, but the insect flew away before he could take more photos. There was no obvious association of the detection with imported risk goods, but the building is across the road from the port where imported vehicles and machinery are de-vanned and cruise ships berth. MPI conducted an inspection of the Britomart precinct in downtown Auckland on 23 February. Vegetation within a 150 m radius of the Rabobank building was inspected but no BMSB or other pentatomids were found. High risk site surveillance inspection of the transects in the area was conducted on 19 January but no BMSB were found. The MPI Border team was informed of the sighting and appeared to be confident that if there were BMSB in the area, they would find them thanks to the increased awareness from targeted campaigns and fact sheets. The investigation was closed despite the uncertainty around the identification of the suspect bug. However, with enhanced awareness among experienced MPI and port staff, any BMSB present are more likely to be found in that area than in any other part of New Zealand.

A Christchurch hotel operations manager reported to MPI that a suspect brown marmorated stink bug *Halyomorpha halys* (BMSB) had been found in one of the rooms. The specimen was collected by an MPI border officer and confirmed as an unmated and non-reproductive BMSB female. Based on the state of ovary development, it had recently come out of overwintering, which was consistent with the state of live bugs found at the border at that time of year. An MPI entomologist and an Incurion Investigator visited the hotel, inspected the site and conducted an investigation. The fifth-floor hotel room in which the insect was found had a fixed window, and the recent occupants were all from New Zealand. However, the hotel receives a large number of visitors each week – for example, it had hosted 44 United States Antarctic Program (USAP) personnel a few days previously in other rooms. It is therefore possible that the insect hitch-hiked on

luggage from overseas. Solitary unmated BMSB on luggage are considered a low risk as the bug has limited opportunity to reproduce. Because of the high impact of BMSB and the limited range of eradication tools available, there has been a passive surveillance campaign to detect BMSB by reporting to the pest and disease hotline. People are usually quick to notice an insect on luggage or against a neutral background (such as a hotel room floor in this instance). Aggregations of BMSB are considered a particularly high risk and are usually expected on imported machinery or vehicles. Since the American personnel appear to be not targeted within the current BMSB awareness programme, material was provided to USAP to distribute among its staff to raise awareness of BMSB. The biosecurity risk in this case was considered low to negligible as site inspection yielded no further signs of BMSB and the original detection was a solitary individual.

A domestic cleaner in New Plymouth found what she suspected to be a live BMSB on the carpet inside a house, and this identification was subsequently confirmed by PHEL (Tamaki). The home owner had recently had relatives from New York State in the US staying for six weeks, and just after they left further visitors from New York stayed overnight. The bug was found on the carpet the day after the second visitor had left. The owner had no contact details for the visitors except via Facebook. Enquiries led to a search of arrival cards for the visitors to find another means of contacting them, but to no avail. An attempt was made to intercept the travellers on their departure to Sydney from Christchurch International Airport, to try and find out where they had stayed during their four nights in the area. All providers of accommodation were also traced and contacted, and information about BMSB was sent to each. There were no further sightings of BMSB and the investigation was closed.

While waterblasting a recently imported vehicle in Auckland, a suspect BMSB was found inside the bumper. The bug was caught and a photo submitted to an Incursion Investigator. Subsequently the PHEL (Tamaki) weekend duty entomologist made an interim identification of *Erthesina fullo*, the yellow spotted stink bug (YSSB). The bug was killed by freezing before being

submitted for a formal identification. It was confirmed as a very old female that was past reproduction and unlikely to have laid any eggs before it was found. The ovaries were degraded and nearly empty, with black material that was interpreted as necrotic oocytes. PHEL reported that it had possibly been starved, as there was no fat in the body at all.

The vehicle had arrived from Japan on a vessel that had discharged vehicles at Ports of Auckland the previous day and was expected next in Wellington. During the 11 days prior to notification the vehicle sat on the wharf for two days before clearance and was then delivered to the owner's home. It was compliance-checked at Mt Maunganui, then went to a panelbeater at Papamoa for paintwork touch ups before return to the owner's home. The owner had worked through a shipping agent, from whom details of the consignment were sought, and provided the contact details for the two commercial properties where the vehicle had work done. MPI border staff distributed YSSB posters and educational materials to enable reporting of new incursions. The contaminated vehicle was transported to the Port of Tauranga treatment facility for fumigation.

Coriander seeds purchased from a bulk bin at an Indian food warehouse were found to have tiny live wasps in them. Specimens received by MPI were identified as *Systole albipennis*, a species not present in New Zealand and a pest of coriander and other Umbelliferae. Commercial production and export of carrot seed from Canterbury has increased significantly in recent years and the risk this species might pose was investigated, including discussion with industry representatives and carrot seed importers/exporters. The investigation concluded that the overall risk was low and was being adequately addressed by the existing Import Health Standard governing coriander seed importation into NZ.

On 13 January 2017 a mechanic working on an imported Hino mini truck contacted MPI regarding a solitary bug found inside the rear cab. The vehicle had been imported on 4 January by his customer. From the emailed picture, the bug was tentatively identified as an exotic species, most likely yellow spotted stink bug *Erthesina fullo*. The MPI border team

was asked to visit the workshop, inspect the vehicle, organise treatment and obtain tracing information. Meanwhile, the notifier was told to keep the vehicle contained at the workshop.

Inspection of the truck was promptly carried out by an MPI border officer, but transporting it for precautionary fumigation presented a new challenge. After extensive discussions with treatment providers to find a suitable carrier, a company agreed to do the job. However, apparently the driveway of the workshop was too narrow for a container to be used for transporting the truck and it was difficult to access the area. Major Auckland transport companies were unable or unwilling to offer the service but eventually two companies jointly completed the task. One company co-ordinated the process, but this was a request they had never had before. This logistical challenge was a good learning exercise to highlight the need for managing future incidents where transport companies are unable to transport infested vehicles for fumigation (especially in a response situation). Further work is needed to develop the best ways to promptly and efficiently treat very large vehicles suspected of harbouring such creatures.

A home gardener in Tauranga found a live suspected BMSB on his kiwifruit vine. The notifier said that the bug was 7 mm long and looked similar to the pictures in the BMSB fact sheet, being brown with white marks on the abdomen and white banding on the antennae. A photo was taken and from this it was confirmed as the brown soldier bug (*Cermatulus nasalis*), which is native to New Zealand.

Plant diseases

The fungus *Phytophthora gregata* was reported by Scion and confirmed by MPI, associated with other *Phytophthora* spp. and other fungi in soil from unimproved pasture adjacent to Waipoua Forest, Northland. Investigation revealed there was no association with kauri trees as originally reported. Soil samples were collected as part of a university student's PhD research. Analysis of earlier *Phytophthora* isolates determined that this was not the first time *P. gregata* had been found in New Zealand. It was first isolated from soil collected in Auckland in 2009 and recorded as "*Phytophthora* sp.". Since *P. gregata* was

described in 2011, molecular analysis has enabled that isolate to be correctly identified and confirmed as still present in New Zealand. Overseas, *P. gregata* has been isolated from soil from pasture and dying trees in Western Australia and Hungary, but pathogenicity testing has not been conducted to determine whether it causes disease in these plants. It was concluded that the biosecurity risk was low and the nature of the detections suggested it was long-established and widespread. No further action was considered necessary.

The Timaru District Council reported an elm tree (*Ulmus* sp.) at Otipua in South Canterbury, with tip dieback, some dead branches and many branch regrowths. The bark was well attached and there was no slime, fungi or cavities. There was concern that the symptoms could be caused by Dutch elm disease, caused by *Ophiostoma novo-ulmi*, but on examination SPS Biosecurity Ltd concluded otherwise. No beetle holes were found on affected branches, but this was not unexpected because the European elm bark beetle *Scolytus multistriatus* is restricted to the upper North Island. Branch samples sent to PHEL (Tamaki) tested negative for *O. novo-ulmi*. The fungus *Diaporthe* sp. was isolated from the sample but it was likely an endophyte or secondary pathogen and unlikely to have contributed to the symptoms observed. *Diaporthe* is known to live in healthy plants but can become pathogenic when they are under stress. While the actual species is still undescribed, DNA analysis shows that it has been isolated as an endophyte from asymptomatic Japanese elm trees (*Ulmus davidiana* var. *japonica*). At least seven species of *Diaporthe* have been reported on elm trees internationally and many records are not identified to species level. The fungus is considered unlikely to pose any biosecurity risks.

References

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- Bulman SR, Visnovsky SB, Hall IR, Guerin-Laguette A, Wang Y (2010). Molecular and morphological identification of truffle-producing *Tuber* species in New Zealand. *Mycological Progress* 9, 205–214.

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PEST WATCH: 4 February – 5 May 2017

Biosecurity is about managing risks: protecting New Zealand from exotic pests and diseases that could harm our natural resources and primary industries. MPI's Diagnostic and Surveillance Services (DSS) directorate devotes much of its time to ensuring that new organism records come to its attention, and to following up as appropriate.

This information was collected from 4 February to 5 May 2017. The plant information is held in the MPI Plant Pest Information Network (PPIN) database. Wherever possible, common names have been included. Records in this format were previously published in the now discontinued magazine *Biosecurity*.

To report suspect new pests and diseases to MPI phone 0800 80 99 66.

Validated new to New Zealand reports

Type	Organism	Host	Location	Submitted by	Comments
fungus	<i>Phaeophleospora eucalypticola</i> no common name	<i>Metrosideros excelsa</i> pōhutukawa	Auckland	PHEL (General Surveillance)	A newly described species, likely a weak or secondary pathogen.
fungus	<i>Ramichloridium punctatum</i> no common name	associated with <i>Polyaspis floccosa</i> flax scale	Auckland	Landcare Research (General Surveillance)	A newly described species. Not a known insect pathogen. Likely to have been present in New Zealand for many years.
insect	<i>Wahlgreniella nervata</i> aphid	<i>Arbutus unedo</i> strawberry tree	Dunedin	SCION / PHEL (High Risk Site Survey)	
spider	<i>Cyclosa fuliginata</i> no common name		Bay of Plenty	PHEL (General Surveillance)	Native to Australia. Likely present in New Zealand for at least 15 years.
virus	<i>Clover yellow mosaic virus</i> CIYMV	<i>Trifolium repens</i> white clover	Auckland	PHEL (General Surveillance)	Likely to be widespread throughout New Zealand in clover.
virus	<i>Persea americana alphaendornavirus</i> 1 PaEV	<i>Persea americana</i> avocado	Northland	PHEL (General Surveillance)	PaEV is symptomless on avocado.

If you have any enquiries regarding this information please contact surveillance@mpi.govt.nz



Veterinary Diagnostic Laboratories

GRIBBLES VETERINARY PATHOLOGY

- **AUCKLAND**
Courier: 37–41 Carbine Road, Mount Wellington, Auckland 1060
Postal: PO Box 12049, Penrose, Auckland 1642
Tel: 09 574 4701 Fax: 09 574 5304
- **HAMILTON**
Courier: 57 Sunshine Ave, Hamilton 3240
Postal: PO Box 195, Hamilton 3240
Tel: 07 850 0777 Fax: 07 850 0770
- **PALMERSTON NORTH**
Courier: 840 Tremaine Avenue, Palmerston North 4440
Postal: PO Box 536, Palmerston North 4440
Tel: 06 356 7100 Fax: 06 357 1904
- **CHRISTCHURCH**
Courier: 7 Halkett Street, Christchurch 8140
Postal: PO Box 3866, Christchurch 8140
Tel: 03 379 9484 Fax: 03 379 9485
- **DUNEDIN**
Courier: Invermay Research Centre, Block A, Puddle Alley, Mosgiel, Dunedin 9053
Postal: PO Box 371, Dunedin 9053
Tel: 03 489 4600 Fax: 03 489 8576

To report suspected exotic land, freshwater and marine pests, or exotic diseases in plants or animals, call:

0800 80 99 66

Investigation and Diagnostic Centre –
Wallaceville
66 Ward Street
Upper Hutt
Tel: 04 526 5600

Investigation and Diagnostic Centre –
Tamaki
231 Morrin Road
St Johns
Auckland
Tel: 09 909 3568

Investigation and Diagnostic Centre –
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14 Sir William Pickering Drive
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Tel: 03 943 3209

NEW ZEALAND VETERINARY PATHOLOGY

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Courier: Cnr Anglesea and Knox Streets, Hamilton
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